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CALORIMETRY IN MEDICINE

WILLIAM S. McCANN

From the Medical Clinic of the Johns Hopkins Hospital

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INTRODUCTION

In the Aphorisms of Hippocrates occur the following interesting statements:

The aged endure fasting most easily; next adults; next young persons, and least of all children, especially such as are most lively.

Growing bodies have the most innate heat; they therefore require the most nourishment, and if they have it not they waste. In the aged there is little heat, and therefore they require little fuel, for it would be extinguished by much. Similarly fevers in the aged are not so acute because their bodies are cold. (Charles Singer (274).)

After more than twenty-three centuries the correctness of the observations of the Father of Medicine is attested by numerous measurements of this innate heat, which the Greeks recognized as a fundamental condition of life.

The foundations of our modern knowledge of the processes involved in the production of animal heat were laid by Lavoisier when he discovered that the amount of ice melted by a guinea-pig parallels the amount of carbon dioxide given off by the animal. He drew the analogy between the emission of heat, the absorption of oxygen and the production of carbon dioxide by a burning candle and by a living animal, and recognized the identity of the processes involved. The plan was broadly conceived by Lavoisier and the foundations firmly laid.

The structure which has been erected on the work of this great Frenchman, though still incomplete, has reached enormous proportions. The history of its development has been admirably told by Lusk. Except for the mention of a few of the principal events this review will not be concerned with the history of normal metabolism. The discovery of the mechanical equivalent of heat by Joule in 1842, of the law of conservation of energy in 1845 by Mayer and Helmholtz, studies by Liebig of the composition of foods and of the tissues into which they are converted, and the final correlation of these discoveries in the masterly work of Carl Voit and Pettenkofer; these events mark the principal epochs in the history of calorimetry.

Curiosity as to the significance for medicine of this new knowledge was evinced very early. The respiratory exchange of phthisical patients was studied by Nysten (228) in Paris in 1811. Regnault and Riesel (249) about 1850 expressed a desire to install suitable apparatus in the hospitals of Paris for the study of the respiration of man in pathological states. Among the earliest studies of Pettenkofer and Voit (241) was that of a man with leukemia. These sporadic efforts yielded little or nothing to medicine because of their prematurity. Before one could measure or detect the abnormalities of the metabolism in disease it was necessary to have a clear definition of the nature and limits of magnitude of the normal metabolic processes.

It is within only the most recent years that the definition of normal values in metabolism has been sufficiently clear to permit the widest use of calorimetry in medicine. The time is now ripe for the practitioner to sum up the knowledge in this field in order to see how he may use it.

Much important information is available concerning the quantitative changes in the heat production in disease. Fever is an every day problem of the physician and calorimetry has much to offer in knowledge of the regulation of temperature. Valuable contributions have been made concerning the effects of drugs and of the secretions of ductless glands, as for instance, in exophthalmic goitre. The treatment of myxedema may be accurately controlled as never before.

The interrelationships of the metabolism with the functional demands on the heart and lungs are of the utmost importance to the practitioner of medicine. Because the demands of the metabolism determine the volume output of the heart and the rate of ventilation of the pulmonary alveoli the physician should know something of the metabolic cost of muscular effort, the mechanical efficiency of his patient. If he understands these things failure of the heart in exophthalmic goitre will lose much of its mystery for him. The proper dietaries in various diseases, long a matter of guess work or subject to individual whims or fads, may now be prescribed with definite knowledge. Most important of all, the energy demands of growing children may be definitely determined. No less fascinating or important is the view unfolded by indirect calorimetry of the intermediary processes of metabolism in a disease such as diabetes.

So far nothing has been said of the diagnostic use of calorimetry, though this use is the chief one by which it is known to the profession at large. In the writer's opinion the measurement of metabolism for diagnosis is the least valuable of all the contributions of calorimetry to the clinic, the most abused and the most dangerous.

Apparatus and methods

It is not considered to be within the scope of this article to enter into a description of the numerous forms of apparatus used in animal or human calorimetry. As sources of such information the reader is referred to articles by Murlin (218), Lusk (166), Carpenter (59),

and Krogh (151a). In general three methods have been employed: (a) direct calorimetry; (b) respiration calorimeters; (c) indirect calorimetry.

Direct calorimetry has been carried out by the use of apparatus such as that of Reichert (250), Ott (230), Paschutin (156) and others (80). Concerning the Paschutin type of calorimeter very little has appeared in the English, German or French literature. A considerable bibliography of the Russian literature of calorimetry has been compiled for the writer by Dr. William A. Perlzweig.

Direct calorimetry involves the physical measurement of heat eliminated from the body by means of radiation, conduction, convection, and the vaporization of water from the skin and lungs. It must take into account corrections for heat stored in the body if the body temperature rises during the period of observation: a correction of positive sign, or a similar correction of negative sign if the body temperature falls. For these corrections the specific heat of the body should be known accurately. The heat production is the algebraic sum of the heat eliminated and the correction for change in body temperature.

Another phase of direct calorimetry is that of determination of the heat of combustion of foodstuffs burned in a bomb calorimeter. (Riche (255).) If the heat of combustion of 1 gram of pure dextrose be determined directly, and if from the chemical formula for dextrose a calculation be made of the amount of oxygen required to completely burn 1 gram of dextrose to CO_2 and H_2O , then one may calculate the heat value of any given quantity of oxygen used in oxidation of dextrose. Similar calculations have been made for the other foodstuffs, but in the case of protein a deduction is made of the heat value of end products of protein metabolism found in the urine and feces. A. V. Hill, Meyerhof and others have used minute and very accurate differential calorimeters, which are capable of measuring the heat of small isolated muscles, ova, small animals, and even of the heat produced in the agglutination of bacteria by an immune serum. (Bayne-Jones.)

Indirect calorimetry is the method of calculating heat production by the measurement of the carbon dioxide and oxygen exchanged in the

respiration. The validity of the method has been established beyond a doubt by the simultaneous measurement of heat by physical means, and of the gas exchange in a respiration calorimeter. Such calorimeters exist in this country in the laboratories of Graham Lusk, of the Russell Sage Institute of Pathology in Bellevue Hospital under the direction of DuBois, at the Nutrition Laboratory of the Carnegie Institution directed by F. G. Benedict, in the department of Agriculture in Washington, and at Pennsylvania State College. Abroad there is the respiration calorimeter of Rubner in Berlin, the first successful apparatus of its kind. All of these employ the principles of Regnault and Riesel for the measurement of respired gases.

DuBois (95) recorded the accuracy of the Russell Sage Calorimeter as follows:

In observations lasting three or four hours the heat production, carbon dioxide elimination and oxygen consumption, as determined by alcohol and electrical tests can be measured with an average error of 0.9, 0.6, and 1.6 per cent, respectively. Such a comparison of the direct and indirect methods establishes the validity of indirect calorimetry beyond a doubt, so that is possible by the use of much simpler apparatus to measure the respiratory exchange alone as a basis for calculating the energy transformations of life.

For the principles of the calculation of the heat production indirectly from the respiratory exchange and urine nitrogen excretion the reader is referred to Zuntz and Schumburg (304), Lusk (166), Benedict and Carpenter (25), and other sources (95, 151). For the complete and accurate calculation of heat production the measurement of both respiratory gases is essential, because the heat value of a unit quantity of either carbon dioxide or oxygen varies with the nature of the foodstuff being oxidized. When fat is burned the caloric value of 1 liter of oxygen is 4.686, whereas when carbohydrate is oxidized 1 liter of oxygen is equivalent to 5.047 calories. Thus the heat generated as calculated from the oxygen consumed or carbon dioxide excreted varies with the non-protein respiratory quotient, which normally lies between the limits of 0.71 (fat) and 1.0 (carbohydrate).

Recently numerous forms of simple apparatus for clinical use have been devised (35). For most of these only one of the respiratory gases is measured. Usually when a single gas is the basis of calculation the one selected is oxygen, because the absorption of oxygen is less affected by abnormalities of respiration than is carbon dioxide. This latter gas exists in such labile combination with bases in the body fluids that overventilation of the lungs will result in an excretion of CO_2 at a rate greater than the actual production of the gas by the oxidations of the body. If overventilation occurs during a period of observation with any apparatus which determines CO_2 alone an erroneous measurement of the heat production will be obtained, which will be much higher than the true value.

In all methods of measuring the respiratory exchange there are inherent difficulties of great magnitude. If one gas alone is measured it is much more difficult to detect an error. If both gases are determined the measurement of one checks the other. In the hands of well trained workers reliable results may be obtained with simple apparatus. The great danger arises from the results of poorly trained or careless observers, who neglect to take adequate precautions to insure the accuracy of their determinations. To base a diagnosis upon the results of such metabolism tests, in which innumerable sources of error may remain undetected, is an abuse of science, an insult to the clinical art, and a crime against the patient.

Total energy requirements in disease. Physiologists have made available for the economist data of the utmost importance, upon which depend the solution of the problems of food supply so vital to a nation in the grip of modern warfare. These problems are becoming steadily more significant for a world at peace as the growth of population makes it approach the limit which the earth is capable of supporting. The same data, which are so useful to the economist, afford points of departure for the investigation of the energy requirements in disease. Such investigations determine not only the dietary needs in sickness, but furnish a more complete understanding of many of the faulty mechanisms of the diseased organism.

In a study of the effect of any given disease upon the total energy requirement it is necessary to have some means of comparing that of

individuals suffering with that disease with that of normal individuals of the same size, shape, age and sex. For this comparison it has been found that the determination of the *basal metabolism* serves best. This furnishes a measure of the overhead cost of maintenance of the individual. In addition to the "overhead" it is necessary to know the effect of food on the energy transformations, the *specific dynamic* action of food, and what may be the cost to the individual of the performance of muscular work, a cost which depends upon the *mechanical efficiency* of the individual.

PART I. BASAL METABOLISM

The term *basal metabolism* was introduced into English by Lusk as the English equivalent of the German "Grundumsatz." It was originally intended to mean the metabolism of an individual at the lowest ebb of functional activity. However, as the minimal heat production probably occurs during sleep in the early morning hours, in practice measurements have come to be made at a more convenient time of day on subjects who are awake. The *standard basal metabolism* is the heat production of an individual at complete muscular rest in the "nuchtern" or post absorptive condition (fourteen to sixteen hours after the last ingestion of food), in an external environment of about 33°C. (comfortable clothing). These are the conditions under which most of the measurements have been made upon which normal standards are based.

a. Surface area and the surface area law

From the beginning investigators have sought to find some physical measurements of an individual with which the observed heat production could be correlated in an attempt to predict with accuracy the metabolism of other normal individuals. For this purpose metabolism has been referred chiefly to the body weight and to the skin surface area. The idea that the basal metabolism is proportional to the body surface area is an old one, having been proposed by Sarrus and Rameaux as early as 1838. (Harris and Benedict (118).) It originated in the conception that the heat loss is proportional to

the body surface and that, since the body temperature is kept approximately constant, there must be a causal relationship between the extent of body surface and the heat production of the body.

Almost simultaneously, about 1883, Rubner and Richet presented experimental evidence tending to show that the basal metabolism is proportional to the body surface. Following this an immense amount of confirmatory evidence was brought out in support of the so called "Surface Area Law," not only as applied to man, but to small and large animals of all kinds. (E. Voit; Armsby, Fries, and Braman (118.))

The study of the applicability of the surface area law made it necessary to have an accurate means of determining the area of the skin surface. For adults the most commonly used formula for this purpose was that of Meeh, which stated that body surface was proportional to the two-thirds power of the weight. Other formulae like those of Lissauer and of Rubner and Heubner which differ from Meeh's only in the constant term, have been used for the surface area of children. Howland and Dana (133) have likewise developed a formula from the data of Meeh's original measurements. D. DuBois and E. F. DuBois (72) after making a series of the most careful and painstaking casts of the body, weighing the paper patterns, were able to compute the skin area from a series of measurements of the body by dividing it up into geometrical figures. This work was known as the Linear Formula, which is the most accurate means of determining body surface, and one which should still be used for very obese persons or for individuals of extremely abnormal shape. It has been found to hold for children over two years of age by Sawyer, Stone and DuBois (265). The average error for the *Linear Formula* is somewhat under 2 per cent. It is a time consuming procedure to make all of the required measurements.

Later DuBois and DuBois (73) devised an easier though slightly less accurate means of predicting surface area from the height and weight. This formula is as follows:

$$A = \text{weight}^{0.425} \times \text{height}^{0.725} \times 71.84$$

where A = area in square centimeters the weight being in kilograms

and the height in centimeters. This formula has been found by DuBois to be accurate within the limits of ± 5 per cent.

Following this Benedict (34) in 1916 described a photographic method of determining surface area. By this method the subjects are photographed in suitable positions and the photographs are measured by means of a planimeter. The area of the body determined in this way agrees closely with the results obtained by the DuBois height-weight formula.

These methods have shown how totally inadequate was the formula of Meeh, with which the average error is 16 per cent, with a maximal variation of 36 per cent. In view of this fact it seems highly improbable that the Lissauer formula for the surface area of children is any more accurate than that of Meeh to which it is entirely similar. (Meeh's formula is $A = 12.3 \times \sqrt[3]{w^2}$ Lissauer's $A = 10.3 \times \sqrt[3]{w^2}$). Benedict and Talbot (38) found that Lissauer's formula could be used for obtaining the surface area of children provided that the value of the constant term was varied with different weights.

Faber and Melcher (86) have tested the DuBois height-weight formula in the measurement of area of infants under two years of age. In a series of 100 newborn babies, the area could be computed from a slightly modified height-weight formula with an average accuracy of ± 2.5 per cent. The modified formula is $A = \text{weight}^{0.425} \times \text{height}^{0.725} \times 78.50$.

b. Normal standards for the basal metabolism of adults

As work was done with the more accurate measures of surface area it became apparent that the original statement of the Rubner-Richet law would require modification, for it was found that age and sex had distinct effects upon the metabolism of man. Aub and DuBois (10) reviewed the available data on normal subjects up to 1917 and from these came to the conclusion that age and sex should be taken into account. They proposed a tentative series of standards for the average heat production per square meter of skin area per hour for normal individuals from fourteen to eighty years of age. From these average figures the normal metabolism was found to vary between the limits of ± 10 per cent.

Standard basal metabolism (Aub and DuBois (10))

Calories per square meter per hour. Area by height-weight formula

AGE	MALES	FEMALES
<i>years</i>		
14-16	46.0	43.0
16-18	43.0	40.0
18-20	41.0	38.0
20-30	39.5	37.0
30-40	39.5	36.5
40-50	38.5	36.0
50-60	37.5	35.0
60-70	36.5	34.0
70-80	35.5	33.0

Aub and DuBois (10) recognized the probability that these standards would have to be modified from time to time as a larger series of normal individuals were studied. These standards have been adopted in practically all American clinics. From various clinics have come many data confirming their validity, for instance the results of the study of persons of markedly abnormal shape, such as patients with simple obesity uncomplicated by pituitary disease studied by Means (201, 202), or dwarfs and legless men studied by DuBois and Aub (11). From a series of 127 normals Boothby and Sandiford (51) have found 92 per cent to have a metabolism within ± 10 per cent of that predicted by the Aub and DuBois standards and that 99.3 per cent were within ± 15 per cent.

Among the chief opponents of the surface area law is Dr. F. G. Benedict of the Carnegie Nutrition Laboratory in Boston. In 1915 Benedict (33) concluded "that the metabolism or heat output of the human body, even at rest does not depend on Newton's law of cooling, and is, therefore, not proportional to the body surface." It is quite obvious that the surface area law might hold, even though the theoretical explanations of the law were invalidated.

Harris and Benedict (118) have given an extensive critique of the surface area law in which many valuable points are brought out. Their first attack is on Newton's law of cooling, which they consider to have very limited applicability. It is furthermore a law of the rate of cooling in relation to the difference in temperature. The

change in heat production at different environmental temperatures is not proportional to the change in temperature. The nature of the integument and the ability of the body to control the distribution of blood flowing through the skin and subcutaneous tissues, as well as the secretion of sweat, undoubtedly affect the rate of heat loss at different environmental temperatures so that Newton's law can not be made to apply as it would in the case of a lifeless body. This objection of Harris and Benedict to the law of cooling can not destroy the surface area law, however, if actual measurements of surface area and basal heat production show them to be proportional to each other.

Again Harris and Benedict (118) point out that during inanition the decrease in heat production is out of all proportion to the decrease in weight or in body surface. Also, a dog confined to the laboratory for a period without loss of weight or of surface area showed a decreased metabolism. These are important points in that they emphasize the importance of interpreting metabolism tests in the light of the nutritive condition of the subject, nevertheless the writer can not regard them as valid objections to the surface area law because the subjects of inanition experiments can no longer be regarded as normal. Starvation partial or complete if persisted in will eventually cause death. It is obvious that a man can be maintained in a normal state only when he is receiving sufficient food to fulfill his requirements. The surface area law applies to the metabolism of *normal* individuals.

Harris and Benedict (118) made an extensive statistical study of the relative values of body weight and body surface as a basis for prediction of the heat production of a subject. The average deviations with regard to sign of the predicted from the actually observed values, and the average deviations without regard to sign showed that the mean heat per unit of body surface was a better basis of prediction than the mean heat per unit of weight. Similar results in favor of body surface were obtained when the bases of prediction were tested by the square root of the mean square deviation of the predicted from the observed values. Likewise when prediction was made by means of linear regression equations a further advantage of body surface was found. These authors conclude that with the best methods of

calculation the superiority of body surface over body weight as a basis of prediction is not very large.

Harris and Benedict then set themselves to the construction of multiple prediction tables by biometric formulas involving no assumption concerning the derivation of surface area but based on direct physical measurements. The equations based on data of observation of 136 normal adult men and 103 normal adult women are as follows:

$$\begin{aligned} \text{For men.....} h &= 66.4730 + 13.7516w + 5.0033s - 6.7550a \\ \text{For women.....} h &= 655.0955 + 9.5634w + 1.8496s - 4.6756a \end{aligned}$$

where h = total heat production per twenty-four hours in calories, w = weight in kilograms, s = stature in centimeters, and a = age in years. The evaluation of these equations, which are used in the calculation of the theoretical heat production for any individual, requires merely the substitution of the actually measured weight, stature, and age. The prediction of metabolism from birth to puberty is left for more complete discussion later when another publication by Benedict and Talbot (38) will be considered.

Criticism of the work of Harris and Benedict has not been lacking. Boothby and Sandiford (51) have made a comparison of the DuBois (10) and the Harris and Benedict (118) normal standards for the estimation of the basal metabolic rate. In a series of 404 determinations there was an average difference of 6.5 per cent in the calculations based upon the two standards. They have calculated the surface area of subjects whose heat production was predicted by the Harris and Benedict tables by dividing the predicted heat production by the corresponding prediction of DuBois and Aub for the heat production per square meter per diem. When the surface areas obtained in this way are plotted graphically on the same coordinates with the surface area determined by the DuBois height-weight chart, the parallelism is striking, and the Harris and Benedict prediction tables have thus been utilized to strengthen the contention of Lusk and DuBois that the basal heat production, for a given age and sex, is proportional to the surface area. The chief discrepancies between the two methods lie in the difference in values attributed to the factors for age and sex. "Harris and Benedict, in their correlation formula, assume that a small subject will show more than twice the percentage

decrease in heat production for advancing age than a large subject, while DuBois assumes that age affects alike both small and large people." (51) "Harris and Benedict assume a reversed action for sex, depending on the size of the subject, by predicting first that large men have a greater heat production than similar sized women and second that small women have a greater heat production than small men." (51) Boothby and Sandiford conclude that the Aub and DuBois normal standards give the best method available at present for predicting the normal heat production, until Harris and Benedict prove that the effect of age varies with size, or that the effect of sex is different in small and large women.

Dreyer (71) has likewise proposed several formulae for the prediction of metabolism. These are based on age, sex, and one other factor, either weight, trunk length or circumference of thorax, the constants being derived from the Harris and Benedict data. The formulae are as follows:

W = net body weight in grams
 C = calories per 4 hours
 A = age in years
 Ch = circumference of chest in cm.
 λ = trunk length in cm.

- (1) $\frac{W^n}{C \times A^{0.1333}} = K_1$ where n = approximately 0.5, K_1 for females = 0.1127 and K_1 for males = 0.1015
- (2) $\frac{\lambda^n}{C \times A^{0.1333}} = K_2$ where $n = \frac{3}{2}$, K_2 for males = 1.567, and K_2 for females = 1.597
- (3) $\frac{Ch^n}{C \times A^{0.1333}} = K_3$ where $n = \frac{3}{2}$, K_3 for males = 1.37, and K_3 for females = 1.76
- (4) $\frac{W^n}{\lambda} = K_4$ where $n = \frac{1}{3}$, K_4 for males = 0.319, and K_4 for females = 0.313
- (5) $\frac{W^n}{Ch} = K_5$ where $n = \frac{1}{3}$, K_5 for males = 0.662, and K_5 for females = 0.30213

Means and Woodwell (208) have compared the accuracy of prediction of the basal metabolism of normal men by the DuBois height-weight surface area method (10), the Harris and Benedict (118) multiple prediction tables and by the Dreyer (71) body weight formula. The average deviations were found to be essentially the

same by each method, though the DuBois deviations tended to be about 2 per cent lower than either of the others. With six obese subjects the Harris and Benedict tables gave a slightly closer prediction than the other two formulae, though the differences by the latter methods were not significant. In abnormal subjects the deviations by the three methods were essentially parallel, though on the average the Harris and Benedict deviation tended to be about 6 per cent higher, and the Dreyer deviation about 7 per cent higher than the DuBois.

Means and Woodwell (208) suggested a slight modification of the DuBois standards (10), by subtracting from 1.8 to 0.6 calories per square meter per hour, which would abolish the differences in prediction by the three methods. They were careful to state that this would offer no material advantage, but since the DuBois method had been so long in use it was considered preferable to continue its use in the interest of uniformity. Nevertheless, the Sanborn Company in a book on Basal Metabolism has given a table of modified standards of Aub and DuBois, under the name of these authors and without their sanction.

c. Normal standards for the basal metabolism of children

The problem of finding the normal standards of basal metabolism in early life is a very difficult one. The most recent publication dealing with the whole subject of basal heat production from birth to puberty is that of Benedict and Talbot (38). These authors review the literature thoroughly. Many of the children previously studied had not been observed under conditions in which the basal metabolism could be ascertained. Even in their own series it is probable that the measured heat production of infants was 8 to 15 per cent above the true basal level owing to the effect of food. The restlessness and crying of unfed infants would have introduced a far greater error. The data from observations of a large number of children have been elaborately correlated in numerous graphs. The mean lines represented "the resultant personal impressions of five skilled workers with metabolism curves." From the average curves of heat production per unit of body weight and per unit of body surface a test was made of the accuracy of prediction of the metabolism.

The percentage deviations of all boys on the basis of weight was ± 7.4 per cent and on the basis of surface was ± 7.5 per cent. Above the weight of 10 kgm. the prediction was more strikingly in favor of weight as the basis. The prediction of metabolism of girls was somewhat less accurate than that of boys. The scattering of points on the graphs for both boys and girls was rather more marked than in the case of adults when heat production is referred to surface area. For instance, with a surface area of 1.3 square meters one boy produced 1096 calories *per diem*, another 1401, the difference being 28 per cent; from 0.80 to 0.85 square meters the variations were from 716 to 984 calories, about 37 per cent; and at 0.53 square meters the variation from 456 to 684 calories amounted to about 50 per cent.

The correlations of body surface referred to weight and of body surface referred to height show remarkably smooth curves, which are almost straight lines. This speaks for a remarkable uniformity in the normals selected and indicates that with normal children of a given weight that the height varies little, and *vice versa*, with normal children of a given height the weight varies but little.

The surface area measurements of Benedict and Talbot were made by the DuBois linear formula (72) (265) in nearly all cases, except in some of the smaller infants. For these the Lissauer formula was used. For the larger children these authors found that the Lissauer formula gave very good results when compared with the Linear formula, provided the K of the Lissauer formula were modified for different age and sex groups as follows:

Values of K in the formula $K \sqrt[3]{W^2}$ for computing body surface area

BOYS		GIRLS	
Weight	K	Weight	K
Up to 6 kgm.	10.0	Up 6 to 10 kgm.	10.1
6 to 15 kgm.	10.6	6 to 10 kgm.	10.6
15 to 25 kgm.	11.2	10 to 20 kgm.	10.8
25 to 40 kgm.	11.5	20 to 40 kgm.	11.1

Faber and Melcher (86) have proposed the use of a modification of the DuBois Height Weight Formula for the prediction of surface area of

new born infants. This formula differs from the original DuBois formula only in the value of the constant term.

$$A = \text{weight}^{0.425} \times \text{height}^{0.725} \times 78.50$$

In a series of measurements of 100 new born infants it was found to have an accuracy of ± 2.5 per cent as compared with the area determined by the DuBois Linear Formula.

Benedict and Talbot (38) have prepared a table of standards for the average heat production of normal boys and girls ranging in weight from 2.5 to 38 kgm. This table is reproduced in part as follows:

*Basal heat production of boys and girls per 24 hours from body weight**

BODY WEIGHT NET	PREDICTED HEAT		BODY WEIGHT NET	PREDICTED HEAT	
	Boys	Girls		Boys	Girls
<i>kgm.</i>	<i>calories</i>	<i>calories</i>	<i>kgm.</i>	<i>calories</i>	<i>calories</i>
2.5	115	110	16	755	710
3.0	150	150	17	780	735
3.5	180	185	18	805	760
4.0	210	220	19	830	780
5.0	270	285	20	860	805
6.0	330	350	21	885	830
7.0	390	405	22	910	855
8.0	445	460	24	965	900
9.0	495	500	26	1020	950
10.0	545	540	28	1070	1000
11.0	590	580	30	1115	1045
12.0	625	610	32	1160	1090
13.0	660	640	34	1200	
14.0	695	665	36	1240	
15.0	725	690	38	1275	

* From Benedict and Talbot (38).

These standards are the best at present available. Our knowledge of the metabolism in the prepubescent state must still be regarded as incomplete because of conflicting evidence from most reliable sources. DuBois (74) studied 8 normal boys 12 or 13 years old. Though they were very quiet their metabolism was 25 per cent higher than the normal adult value per square meter of body surface. This increase in metabolism was attributed by DuBois to the greatly accelerated

growth just preceding puberty. These same boys were studied again two years later by Olmstead, Barr and DuBois (229) who found that the heat production per unit of body surface had decreased 13 per cent, so that at the ages of 14 to 15 it was only 11 per cent above the adult level. In the three youngest boys the metabolism during the twelfth year was actually greater in calories produced than during the fourteenth year, although the boys showed a gain in weight of between 35 and 50 per cent.

The findings of Benedict and Talbot (38) do not show any evidence of a greatly accelerated metabolism in the period just before the onset of puberty, nor of a fall in metabolism during adolescence. In the face of contradictory evidence from the laboratories of two such workers as Benedict and DuBois no conclusions may be drawn until much more evidence is available.

The metabolism of *premature and undersized infants* has been studied by Talbot and Sisson (284) and by Murlin and Marsh (222). The former found an average heat production of 597.3 calories per square meter per diem in a series of 22 premature infants varying in age from 3 days to 3 months, or about 25 calories per square meter per hour. Six of these infants, studied in the first eleven days, had an extraordinarily low metabolism ranging from about 330 to 500 calories per square meter per diem. These results are in substantial agreement with those of Murlin and Marsh (222), who found an average of 24.6 calories per square meter (Lissauer) per hour in a series of 10 new born infants all weighing less than 5 pounds and 5 ounces. Only 6 of the 10 survived and these had an average heat production of 25.8 calories per square meter per hour. One of the infants, who did not survive, was a case of *sclerema neonatorum*, with a metabolism of 24.06 calories per square meter per hour. This is the only record of the metabolism in this disease.

d. The basal metabolism in pregnancy

Carpenter and Murlin (58) investigated the heat production of 3 pregnant women just before and just after parturition. The mothers showed a loss during parturition of about 20 per cent of the *ante-partum* weight, and a reduction of about 14 per cent in heat production. In two of the three cases the metabolism of the child was equal

to the difference in the mother's metabolism. Unfortunately the calculations relative to surface area were all based on the unsatisfactory Meeh formula. The relation of the metabolism of the mothers to body weight was almost identical before and after parturition. These results agree with those of Hasselbalch (119). In Hasselbalch's case the oxygen consumption during the last month of pregnancy was 17 per cent greater than it was six weeks *postpartum*. In an observation of Magnus Levy's (quoted by Lusk) (166) the oxygen consumption of a pregnant woman increased by 27 per cent between the first and ninth month.

Murlin (215) studied the gas exchange of a female dog during two pregnancies and during sexual rest. In each pregnancy the extra heat production of the mother was proportional to the weight of the litter at birth.

Baer (18) observed the basal metabolism of 44 normal women late in pregnancy. He found the oxygen consumption per unit of surface area to be 33 to 35 per cent above that of the nonpregnant woman. This increase was not believed to be due entirely to the fetus because 3 days after delivery the average metabolism was still 15 per cent above normal, reaching the normal basal level from 7 to 10 days *postpartum*. Baer found that death of the fetus could be detected by the finding of a basal metabolism which was below the average of this series in a woman otherwise normal. It is very unfortunate that Baer failed to give more of the actual experimental data in his publication, and that a more reliable form of apparatus was not used.

Root and Root (260) report the measurements of the basal metabolism from the fifteenth week of pregnancy to the eighth week *postpartum*. The subject was a perfectly normal primipara. The basal metabolism in the fourth month was normal, but from that time on increased steadily up to eleven days before delivery, when the total calories were 23 per cent greater than in the fourth month. The calories per unit of body weight were found to have increased 7.6 per cent in the same period. Following delivery though the subject's weight remained the same the heat production per unit of weight was 9.6 per cent below that of the fourth month of pregnancy. Even though the subject herself was perfectly quiet considerable variations from period to period were noted on the same date. It was suggested

that these unexplained variations might have been due to fetal movements.

These authors have very wisely pointed out the necessity of developing normal standards for the basal metabolism of pregnancy from the study of normal pregnant women.

e. Effects of climate and other environmental factors on the basal metabolism

So far this discussion has dealt with the basal metabolism of normal subjects in the temperate zone only. Whether the same standards for normal heat production are applicable in the *tropics* or in the arctic regions has not yet been determined. That the basal metabolism of white men in the tropics is lower than in temperate climates is maintained by de Almeida (3). This author studied ten white men between the ages of twenty-three and forty years, who resided between the latitudes of $1^{\circ} 37' \text{ S.}$ and $22^{\circ} 54' \text{ S.}$, in a region with a mean temperature variation from 20.6° to 25.6°C. and a humidity from 78 to 88 per cent. The observations were made at Rio de Janeiro with the Tissot method. The average heat production was found to be 30.3 calories per square meter (height-weight formula) per hour, which is about twenty-three per cent below the DuBois standard. The lowest heat production was 25.7 calories per square meter per hour, the highest 32.4. The variations from average normal were from minus 35 per cent to minus 18 per cent. The higher values were observed in subjects who led active lives, the lowest heat production was that of "a professor who never walked about or exerted himself in any way."

The same author (3) studied ten negroes, males between twenty and forty-four years of age, who showed a mean heat production of 32.86 calories per square meter per hour which is seventeen per cent below the DuBois standards. The lowest heat production among the negroes was 26.3 calories per square meter per hour.

Young (301) reports observations of metabolism of normal white men residing at latitude 19° south. He found that during the hot season of the year the metabolism was greater than in the cooler season, except on certain days in the latter when the rate of cooling was sufficiently great to induce shivering. The explanation given for

the greater metabolism in the hot season is that during the hot moist weather the activities of the subject preceding the observation had led to an elevation of body temperature due to the low rate of cooling.

The studies of Eijkman (84) on the food consumption of white men and Malays living in the tropics do not indicate that the heat production is lower in hot climates. More recently Knipping (145) has arrived at similar conclusions as a result of studies made with a small respiration apparatus.

Lindhard (158) investigated the seasonal variations in respiration and metabolism. In only two of his subjects was the basal metabolism followed throughout the year. With one of the individuals the maximum variation in oxygen consumption throughout the year was four per cent. The other individual showed a progressive rise in oxygen consumption from February to August, falling in September, with some irregularities thereafter associated with change of habits during the holiday season. The variation was 10.5 per cent of the lowest value. These seasonal variations were attributed by Lindhard to the varying intensity of the sunlight. He noted an increase in metabolism during four days following a light bath. The seasonal variation in oxygen consumption is too small to be of significance. The striking thing is the relative constancy of oxygen consumption as compared with the marked seasonal changes in the respiration.

The effect of posture in the metabolism is not negligible. Emmes and Riche (85) found that the heat production was eight per cent greater with the subject sitting up than lying down. Soderstrom, Meyer and DuBois (279) found that a subject reclining in a steamer chair produced 3 per cent less heat than when lying flat in bed. Lusk (166) has given a very excellent review of the literature of the effects of temperature, humidity, wind, light, hot and cold baths, clothing and the effects of electrical currents.

That the basal metabolism is not much changed by ascent to *high altitudes* is shown by the work of Durig and Zuntz (81), and Douglas, Haldane, Henderson and Schneider (69). More recently Schneider (268) observed an individual whose resting metabolism was fifteen per cent greater before acclimatization than it was after this had occurred. No significant change in the basal gas exchange occurred in the subjects of Hasselbalch and Lindhard (120) when placed in a chamber at low oxygen tensions.

f. Nutritive disturbances and basal metabolism

The problem of the underweight individual is an interesting one. Blunt, Nelson and Oleson (43) found that underweight children about fourteen years of age had a higher metabolism than that of a group of normal children of the same age, and that the metabolism was sometimes as high as forty per cent above that read from the curves of Benedict and Talbot (38) and in most cases higher than the highest observed heat production of the children of the same weight from which the curve was drawn.

For infants suffering from severe malnutrition Talbot (283) found that the metabolism per unit of weight and of body surface was likewise increased. His conclusions were based on analysis of his own data, as well as the previous work of Howland (132) and of Murlin and Hoobler (219). The metabolism of infants with malnutrition was not found to be greatly changed until the weight loss reached 20 per cent. As fat is lost there is less insulation against heat loss, and the looseness of the skin probably augments still further the relative increase of body surface as compared with weight. These factors tend to increase heat loss. When the heat loss becomes greater than heat production the body temperature tends to become subnormal. It is of interest that in this paper Talbot (283) practically admits the possibility of a causal relationship between heat loss and heat production.

In Talbot's analysis of these cases of malnutrition the metabolism per kilogram of actual weight is the higher the greater the degree to which the subject was underweight. The heat production per kilogram of normal weight lies within normal limits for many of his subjects, and below normal limits for the most severe cases. This finding would indicate that the loss of inert fat alone had occurred in the milder cases, while in the more severe cases there was a reduction in amount of active protoplasm.

The effects of inanition on the metabolism have been recently reviewed by Lusk (173). The surprising fact is brought out that partial inanition produces practically as great a decrease in heat production as does complete starvation. Lusk calculates that Benedict's fasting man (24) lost 16 per cent of his body nitrogen with a

reduction in heat production of 29 per cent, and that Benedict's Squad B (37) lost 3 per cent of their body nitrogen with a reduction of 32 per cent in metabolism due to a reduction of food intake from 4000 to 1375 calories *per diem*. Lusk (173) points out that there appears to be "a biological adaptation to a lowered energy intake, preventing the exhaustion of the reserve of body fat."

When the reserve of body fat is depleted the body is no longer able to spare its tissue proteins. The increased metabolism of protein is associated with an increase in heat production. This is well illustrated in the observation on themselves made by Zuntz and Loewy, who are quoted by Lusk (173). Loewy's metabolism before the war was 726 calories per square meter. Due to inanition it had decreased to 631 calories per square meter *per diem* in 1916. In 1917 Loewy's nitrogen metabolism increased rapidly and at the same time his heat production rose to 738 calories per square meter *per diem*.

These observations of the effect of inanition on the metabolism of previously healthy individuals are important in consideration of inanition occurring as the result of disease. In evaluating the effects of a disease upon the total metabolism the nutritive condition of the patient must be kept in mind.

The adequacy of a diet is not determined by its energy value alone. The amount and quality of the protein constituents are of great importance. However, there have been no studies of the effect on the heat production of man induced by a diet containing only proteins of low biological value. A very few studies have been made relative to the effects of vitamine deficiencies on the metabolism of fowls, and on the results of deficiencies in mineral constituents. These should be fertile fields for further investigation.

Pedotti (238) reported experiments in which the metabolism of rats was measured while they were fed with diets deficient in calcium. A lowered value for the metabolism of these rats was found. It is not clear that this reduction was due to the calcium deficiency or whether it was the result of the ingestion of potassium, by the administration of which it was hoped to deplete the body of calcium.

Ramoino (247) was the first to study the gaseous exchange of pigeons fed on a diet of polished rice. This writer found that the respiratory quotients of these pigeons fell progressively until at the time of

development of the symptoms of polyneuritis quotients from 0.4 to 0.5 were obtained. The lowering of the quotients was due to increase in oxygen consumption rather than to a relative decrease in carbon dioxide production. The abnormal quotients and the variance of the results of this author as compared with the later work of Novarro (227) and of Anderson and Kulp (5) are probably the result of faulty technique.

So far as the writer can discover no study has been made of *avitaminosis* in man. Novarro (227) compared the effects of starvation and of a polished rice diet on the metabolism of doves. During starvation the doves lost weight, developed subnormal body temperatures, and eliminated progressively less heat per square centimeter of body surface. The heat production fell to from 40 to 66 per cent of the normal value, when the body weight had decreased from 23 to 32 per cent. Doves fed on polished rice took less food, lost weight, showed a markedly lowered heat production and a corresponding fall in body temperature.

Novarro (227) found that in the recovery from fasting the metabolism increased above the normal value as the temperature rose to normal, the effect lasting 5 to 6 days. A similar behavior was noted during the recovery from polyneuritis.

Anderson and Kulp (5) have reported quite similar studies during the development of polyneuritis in poultry. There was a loss of appetite and a diminished consumption of food, with a coincident lowering of metabolism amounting in some cases to 40 to 50 per cent. Digestion and assimilation ceased and the respiratory quotients were lower during polyneuritis, but no quotients similar to those of Ramoino were noted. When the fowl recovered from polyneuritis the metabolism rose quickly but the appetite remained poor and the gain in weight was slow.

g. Day to day variations in basal metabolism

Before beginning a discussion of the basal metabolism in disease it is well to consider the extent to which the metabolism of normal individuals may vary from day to day. The number of published reports of repeated observations on the same individual is not large. In the experiments of Lindhard (158) a remarkable constancy of

metabolism was found through a period of a year. Lusk (173) quotes from Zuntz' and Loewy's observations on themselves. Between 1888 and 1910 the metabolism of Zuntz averaged 787 calories per square meter *per diem*, with variations of ± 2.5 per cent. These, however, are observations on trained subjects. In the writer's opinion the chief factor insuring greater constancy of the metabolism of trained subjects, as compared with untrained individuals, is that in the case of a trained subject the conditions of life preceding the observations are usually more rigidly controlled. Even trained subjects residing at a distance from the laboratory are exposed to widely different environmental conditions. In a considerable experience with hospital patients, who were untrained subjects, the writer has been impressed with the small variations in metabolism on repeated observations. These untrained subjects from hospital wards are kept under unusually constant environmental conditions. On the other hand workers like Benedict (33) emphasize the variability of the basal metabolism. When one considers that Benedict's subjects come to the laboratory in the morning from their homes, having been exposed en route to all sorts of varying conditions of weather it is not surprising that considerable variations in basal metabolism should be observed. The same factors must enter into determination of the metabolism of dispensary or office patients, and should be taken into consideration in the interpretation of results. If the reader has any doubts as to the importance of the effects of environmental temperature, humidity, wind, and kind of clothing, he is referred to a very complete discussion of the subject by Lusk, in his *Science of Nutrition*, Chapter IV.

Means (209) emphasizes the importance of the effect of emotional disturbances on the basal metabolism. These disturbances are not uncommon in patients on being first subjected to experimental observation, especially in women and children. DuBois has always emphasized the importance of carrying out a "dummy" experiment on the day preceding the first actual observation in order to allay the fear of the patient regarding an unknown procedure.

Blunt and Dye (43) have reported 216 observations on 17 normal women from 21 to 44 years of age. The daily variations noted were quite large. The greatest difference of maximum above minimum basal

metabolism was 28.8 per cent, and the lowest 7.4 per cent, and the average 13.2 per cent. Only six of the seventeen subjects showed a range lower than 10 per cent. It should be pointed out again that these individuals were subjected to much more variable conditions preceding the observations than is usual with hospital patients, though they are quite comparable to dispensary and office patients. The custom in DuBois' clinic of considering the normal zone of metabolism to lie between the limits of ± 10 per cent of the average (total range of variation 20 per cent) seems justifiable with hospital patients. However, with subjects who are ambulatory it is better to extend this to the limits of ± 15 per cent allowing a maximum variation of 30 per cent between the highest and lowest normal basal metabolism.

Blunt and Dye (43) supply very important information relative to the alleged effect of the menstrual cycle on the metabolism of women. They found that the average of menstrual and intermenstrual observations was the same and that no rhythmical periodic variations in metabolism could be noted. This confirms the findings of Zuntz and of Gephart and DuBois (96), and is at variance with the findings of Snell, Rowntree, and Ford (277), and of Rowe and Eakin (261). The latter did not publish the details of their experiments.

h. The basal metabolism in disease

At the Mayo Clinic studies have been made of the respiratory exchange of an enormous number of hospital patients. Boothby and Sandiford (50) have been able to analyze the results of metabolism determinations upon 8614 subjects, with reference to the variations from the average normal standards. Of these subjects 6197 were patients with thyroid disease. Of the remaining 2417 subjects, 77 per cent gave results which were within the limits of ± 10 per cent of the Aub and DuBois (10) standard values, while 90 per cent were within the limits of ± 15 per cent of average normal (Aub and DuBois). This indicates that there are relatively few diseases, other than those of the thyroid gland, in which the basal heat production varies markedly from normal. Means and Burgess (210) have reported the results of determinations made upon 1000 patients, the majority of whom had, or were suspected of having, thyroid disorders, though

patients with various blood diseases, disorders of other endocrine glands, and various miscellaneous diseases were included. The numerical preponderance of thyroid disease as a cause of abnormal basal metabolism does not justify the assumption that the variations in heat production observed in other diseases are insignificant. The extensive investigations of these authors serve to emphasize the necessity of interpreting metabolism tests only in the light of the most thorough clinical study.

Thyroid disease. Patients with Basedow's disease were observed by Friederich Müller to lose body weight and nitrogen in spite of an intake of food which would have been adequate for normal individuals. From this Müller predicted that the metabolism in this disease would be abnormally high. This prediction was confirmed by Magnus Levy (178) in 1895. This author gave thyroid gland to a normal girl, aged 23 years, height 156 cm., weight 77 kgm. The basal metabolism of this subject before taking thyroid gland was within 1 per cent of the average normal standard of DuBois and Aub (10). On the 19th day of the administration of thyroid extract the oxygen consumption was found to have increased 12 per cent. Magnus Levy (178) then studied three patients with Basedow's disease, finding that their oxygen consumption per kilogram of body weight was much in excess of that of three normal subjects. In 1904 he (182) reported the finding of low metabolism in myxoedema. The results of these studies received abundant confirmation in many clinics. A full appreciation of the importance of these contributions was not possible until normal standards for basal metabolism were made available by the work of DuBois (75, 77). His announcement in 1915 and 1916 of his investigations of the metabolism in thyroid disease was the starting point of tremendous activity in this field. DuBois pointed out the close correlation between the degree of variation from the average normal metabolism and the severity of the clinical manifestations of thyroid disease. The close parallelism made it possible to use the measurement of basal heat production as the most accurate criterion for determining the efficacy of treatment.

The application of this method of study to the numerous cases of thyroid disease at the Mayo Clinic has enabled Plummer and Boothby to make most valuable contributions to the classification of these

disorders. The most recent analysis of their material (about 6200 cases) is given by Boothby and Sandiford (50). *Hyperthyroidism* occurred in patients with exophthalmic goitre, in certain patients with adenomata of the thyroid, in a number of cases of thyroiditis, and in some subjects with malignant tumors of the thyroid. Of the patients with exophthalmic goitre 98 per cent showed variations of more than 10 per cent above the average normal values of DuBois and Aub. Seven per cent were observed to be within the limits of ± 15 per cent of normal. Of patients with thyroid adenomata (2536) only about 56 per cent showed evidences of hyperthyroidism, with the basal metabolism more than 10 per cent above average normal. Slight increases (16 to 20 per cent above average normal) were noted in a small number of patients with colloid goitres, of whom the vast majority proved to have a metabolism within normal limits.

The occurrence of low rates of metabolism in patients with cretinism, spontaneous myxedema, and post operative myxoedema was noted. In a series of 86 cases, 90 per cent of whom had low metabolic rates the clinical diagnosis of hypothyroidism was "questionable." Plummer states that the edema of myxedema becomes recognizable when the basal metabolism is 15 to 17 per cent below average normal. A very excellent correlation between the clinical manifestations and basal metabolism in myxoedema has been made by Sturgis (282).

Plummer, who has compared the phenomena attending the elevation of metabolism by thyroxin with those occurring in the hyperthyroidism of patients with thyroid adenomata, concludes that the condition of such a patient is one of pure hyperthyroidism (244). On the other hand he does not think the phenomena of exophthalmic goitre are those of pure hyperthyroidism. Boothby gives an excellent tabular comparison of the clinical findings and metabolism studies of these two principal groups of patients with hyperthyroidism (49). The striking fact is brought out that when hyperthyroidism occurred under 40 years of age, it was due to exophthalmic goitre in 87 per cent of the cases. The cases with adenoma were in general characterized by the longer duration of goitre before the onset of hyperthyroidism, by the rarity of exophthalmos, absence of palpable thrills or bruits over the thyroid area, and a lower average deviation from average normal metabolism than in the case of exophthalmic goitre.

That all the symptoms of exophthalmic goitre (Graves' Syndrome) cannot be explained by hyperthyroidism is a view held by Kessel, Lieb, and Hyman, who have contributed most valuable studies of this disease (141, 142, 143). Certain "sympathomimetic" symptoms distinguish the Graves' Syndrome from the hyperthyroidism induced by administration of active thyroid preparations. They point out that thyroid hyperplasia and thyroid adenoma may exist for years without at any time causing sympathomimetic symptoms (such as tachycardia, exophthalmos, tremor, sweating, asthenia, diarrhea). These symptoms they believe to be due to a disturbance in the involuntary nervous system. They differentiate from true exophthalmic goitre a group of patients who present the evidences of "autonomic imbalance" whose basal metabolism is normal (141). True exophthalmic goitre is a diagnosis used by these authors only when there is found to be an elevated basal metabolism in patients with these sympathomimetic symptoms. They acknowledge that this constitutes a rather arbitrary division of their cases, and make it clear that they do not regard the changes in the thyroid gland as being causative of the disease, as there are many reasons for believing them to be secondary.

Whatever may be the correct view concerning the etiology and pathogenesis of exophthalmic goitre, these studies of Kessel, Lieb, and Hyman serve to emphasize the existence of a considerable group of individuals who present many of the clinical manifestations of the syndrome described by Graves in 1835, whose basal metabolism may be normal. These patients are recognized by Means as "borderline" cases. Means and Burgess (210) find that "patients with atypical or incomplete evidence of abnormal thyroid function may show normal or abnormal metabolism. The majority show normal metabolism." Judgment concerning the nature of the disorder presented by these borderline patients must be deferred until further studies have been made.

One may now ask, what is the rôle of the basal metabolism determination in the diagnosis of thyroid disease? Means and Burgess (210) state the answer to this question in a very rational way. They conclude that:

1. Patients with an outspoken clinical picture of hyperthyroidism invariably show increased metabolism, and those with definite clinical pictures of hypothyroidism invariably show decreased metabolism. Those with goiters, but no signs or symptoms of abnormal thyroid function, for the most part show normal metabolism.

3. By inference from the indirect evidence we believe that in these borderline thyroid cases, provided that in the first place a true basal rate is secured, and, provided that certain well recognized causes for increased metabolism, such as fever, acromegaly, leukemia, and severe anemia are excluded, the finding of increased basal metabolic rate is strong presumptive evidence of hyperthyroidism. In a similar way, provided that such conditions as starvation, hypopituitarism, and hyposuprarenalism are excluded, a low metabolic rate is strong presumptive evidence of hypothyroidism.

4. To that extent, then, the metabolism test is distinctly useful in differential diagnosis. Like all other laboratory tests it should only be interpreted with due regard to all other clinical and laboratory findings, and with due regard for its limitations and pitfalls.

Metabolism determinations are unsurpassed as an objective means of measuring the severity of hyperthyroidism or hypothyroidism and the response to various forms of treatment. The administration of active thyroid preparations to patients with myxoedema and cretinism can be accurately controlled. As a result of his studies of hypothyroidism Means estimated that doses of from 3 to 4 grains of thyroid extract daily should be ample to bring the metabolism to normal in two or three weeks, and doses of 1 to 2 grains daily should usually be sufficient to keep it there. The requisite dosage of Kendall's thyroxin may be more accurately predicted. Plummer estimates that "the tissues of an average normal man (exclusive of the thyroid) contain 14 mgm. of thyroxin, and that this tends to be exhausted at the rate of 0.75 to 1 mgm. daily. A shift of 1 mgm. thyroxin in the tissues of the body is accompanied by a corresponding rise or fall in basal metabolism of between 2 and 3 per cent. Fourteen milligrams of thyroxin given to a thyroidless individual is not fully exhausted until from the end of the fifth to the eighth week. Two milligrams of thyroxin a day may keep the basal metabolism from 20 to 30 per cent above normal; 3 milligrams a day may hold the basal metabolism 50 per cent above normal" (244, 245).

The following table taken from a publication by Boothby will be of value to those who wish to use thyroxin (49, 245).

Increase in basal metabolic rate from intravenous injection of thyroxin

DIFFERENCE FROM AVERAGE NORMAL METABOLISM	Increase per mgm. 2.8 Average variation 0.7 = 25% Largest + Variation 2.8 = 100% Largest - Variation 1.8 = 64%				Increase per mgm. per square meter. . 4.7 Average variation 1.1 = 23% Largest + Variation 2.9 = 60% Largest - Variation 2.9 = 60%			
	VARIATION IN CALORIGENETIC ACTION DEPENDING ON ORIGINAL LEVEL OF BASAL METABOLISM				VARIATION IN CALORIGENETIC ACTION DEPENDING ON SIZE OF DOSE			
	Average dose	Increase per milligram	Average dose per square meter	Increase per milligram per square meter	Average dose	Increase per milligram	Average dose per square meter	Increase per milligram per square meter
<i>per cent</i>	<i>mgm.</i>	<i>per cent</i>	<i>mgm.</i>	<i>per cent</i>	<i>mgm.</i>	<i>per cent</i>	<i>mgm.</i>	<i>per cent</i>
to -14	8.7	3.3	5.7	5.0	4.8	3.5	3.6	5.0
-15 to -19	9.1	2.3	5.1	3.9	6.9	2.8	5.2	5.0
-20 to -24	7.3	2.9	4.6	4.6	8.8	2.9	7.0	5.2
-25 to -29	10.9	2.8	6.2	5.0	11.2	3.3	9.3	4.8
-30 to -34	14.8	2.9	9.2	4.9	16.0	2.6	11.5	2.7
-35 and below	15.5	3.0	8.6	5.1	24.0	1.6	13.2	3.0

While such a table may be of great service in determining initial doses of the drug for patients, the variations in calorigenetic action are sufficiently marked to make it necessary to control the administration by means of metabolism determinations.

By means of repeated observation of the basal metabolism it has been possible to compare the results of various methods of treatment of hyperthyroidism. DuBois (75) found that mental and physical rest was the surest means of securing a decrease in metabolism. He observed that in three out of four cases ligation of the thyroid arteries caused an increase in metabolism of unknown duration. It lasted for several weeks. The writer has also observed this occurrence. It is not invariable for such an increase to occur after arterial ligation, when a rise occurs it is probably due to rough handling of the thyroid with consequent liberation of an excess of thyroxin. In a study of several cases in which the ligation operation was done by Dr. Emil Holman in the surgical clinic of the Johns Hopkins Hospital no rise in metabolism was observed following operation.

Boothby (49) reported the operative results in 284 cases of exophthalmic goitre and adenoma. Of cases requiring two preliminary ligations and a period of rest followed by thyroidectomy the average metabolism was reduced from 68 to 20 per cent above average normal rate. Of those requiring one ligation followed by thyroidectomy the reduction was from 60 to 17 per cent above normal. For primary thyroidectomies the average variation before operation was +36 per cent and after operation 8 per cent. Reports such as this show that at least temporary relief of hyperthyroidism may be obtained by surgical procedures. The late results of this series of cases has not yet been reported. While undoubtedly many patients obtain permanent benefit from surgical treatment a recurrence of hyperthyroidism is noticed in a certain proportion of the cases of Graves' disease following a period of temporary relief.

Means and Holmes (211) of the Massachusetts General Hospital report a series of observations of the effect of the Roentgen-ray treatment of hyperthyroidism. Their recent series of cases covering a period of three years included 44 patients with exophthalmic goitre and 14 with adenomata. About two-thirds of the patients with exophthalmic goitre either recovered or improved under Roentgen-ray treatment. The remaining third neither improved nor grew worse. Some patients who were not cured were made better operative risks. Only such patients with adenomata who had refused operation were treated with Roentgen-rays.

Richardson (252), also of the Massachusetts General Hospital, reported the results of treatment of 30 cases of exophthalmic goitre treated by subtotal thyroidectomy. Since both the surgically treated and the Roentgen treated cases were in the same hospital, under identical conditions, and studied in the same metabolism laboratory, Richardson's comparison of the two series of cases is of great value. On the basis of the metabolism tests and weight curves he concluded that subtotal thyroidectomy was a more effective form of treatment than the Roentgen-ray treatment. He concedes that the latter treatment produces beneficial results and that in selected cases the Roentgen-ray may be used for a period of 4 months (about 5 treatments). If after 4 months the degree of improvement in general condition and basal metabolic rate do not promise "cure," operation should be undertaken.

One of the difficulties hitherto experienced in deciding on the merits of any form of treatment of exophthalmic goitre has been the absence of any control series of untreated cases. No one had any accurate data regarding the spontaneous course of the disease. Kessel, Lieb and Hyman (143) are making a most valuable contribution to the subject in furnishing a series of carefully studied untreated cases of exophthalmic goitre. Their patients, 38 in number, were simply placed under ideal conditions in a hospital ward. A striking tendency was noted for the metabolism to decrease, and this was more rapid than the average in Means and Aub's Roentgen-ray series (206). On an average, four months after their admission to the hospital these patients of Kessel were restored to economic and social usefulness.

In regard to the treatment of cretinism Talbot and Moriarty (285) believe the determination of the basal metabolism to be of the greatest value. These authors compared the heat production of 10 cretins with the normal curves established by Benedict and Talbot (38) for the metabolism of children per unit of body surface area, and found it was below normal in all their cretins. The doses of thyroid administered were regulated so as to bring the metabolism up to the expected metabolism of normal children of the same age. They believe that a correct diagnosis of cretinism can be made as early as the third month of life. It is strongly recommended that treatment be commenced at the earliest possible date so that proper development may take place. In 1922 Talbot (286) stated that it was still difficult to draw conclusions regarding the normal standards for metabolism of children and issued a warning against the interpretation of studies of pathological metabolism at the present time. The diagnosis of cretinism at any age should never depend upon a metabolism test alone.

The basal metabolism in fever. Many reviews of this interesting subject have been made (102, 148, 149). One of the best, which covers the literature up to 1909, is that of Richter (257). More recently Barbour (19) has dealt with the mechanism of temperature regulation. The most recent review of the calorimetry of fevers is that of DuBois (78), who has been able to make some highly important generalizations as the result of the long series of studies of various

fevers which have been carried out in his clinic during the past twelve years.

The first of these investigations concerned typhoid fever. Coleman and DuBois (63, 64) found the metabolism of typhoid patients to be from 36 to 40 per cent above normal. The level of the basal metabolism did not appear to be changed appreciably if the patients were given the so called "high calorie diet" of Coleman (62), which supplied as much as 170 calories per kilogram of body weight. The basal heat production was found to vary in a curve roughly parallel to the body temperature. At the height of the fever it was usually about 40 per cent above normal, but sometimes as high as 50 per cent. The respiratory quotients were normal, indicating that the intermediary processes of metabolism, were not grossly altered. Toward the last days of fever there was a notable tendency of the respiratory quotients to rise. Direct and indirect calorimetry agreed within 2.2 per cent, a truly remarkable technical feat.

Malarial fever was studied by Barr and DuBois (21.) Except for the period of the chill the level of the heat production was found to be roughly proportional to the body temperature. The chill, at the onset of the malarial paroxysm greatly increased the heat production (200 to 300 per cent). The heat elimination was also slightly increased but not in proportion to the rise in heat production. The net result was a retention of heat within the body and, in consequence, there was a rise of body temperature. The chill having ceased, the metabolism remained elevated above the normal basal level. As the body temperature fell the heat lost greatly exceeded the heat production of the body, which was only slightly above the normal afebrile level. Barr and DuBois pointed out that there are wide differences in the temperature of various parts of the body during fever. They devised a method of calculating the changes in average body temperature based on the estimated specific heat of the human body (about 0.83) and a knowledge of the net gain or loss of heat. The absolute level of this average body temperature could not be ascertained. It was assumed to be about $0.5^{\circ}\text{C}.$ below the rectal temperature. There was a close parallelism between the rectal and the average body temperature, though changes in the course of the former generally preceded those of the latter.

Barr and DuBois quote the classic work of Liebermeister (155) on the malaria paroxysm, and also that of Likhachev and Avroroff (156), who used the Paschutin calorimeter.

The metabolism in tuberculosis was studied by McCann and Barr (190) and later by McCann (191). During periods when the body temperature was normal tuberculous patients were found to have a basal metabolism within normal limits or slightly above normal. As the body temperature rose the metabolism increased, so that in a patient whose rectal temperature was 40°C. (104°F.) the metabolism might be about 30 per cent above the average normal value. The actual heat production of the tuberculous patients was found to be below that which would have been normal before loss of weight occurred. In other words the loss in weight and consequent decrease in body surface area are accompanied by a proportionate decrease in heat production.

Grafe (109) however reported that in afebrile tuberculosis the metabolism might be from 20 to 36 per cent above the average normal of Aub and DuBois (10). This, the writer believes, is an erroneous conclusion. Grafe's patients were kept from 4 to 7 hours in a Jacquet chamber. In such a long period of observation it would require superhuman efforts to remain motionless. Grafe had no means of recording the movements of his patients.

In a study of 15 cases of afebrile pulmonary tuberculosis, 8 of whom had active lesions, Kocher (146) found the basal metabolism varied but a few per cent above to a few per cent below the average normal.

Pierson (242) found marked variations in the basal metabolism of tuberculous women associated with various phases of the menstrual cycle. The experimental results were given only in graphs. The fluctuations in 5 afebrile cases were almost unbelievably large, covering a range of from 30 to 35 per cent. Pierson believes that there is a premenstrual rise in metabolism with a post menstrual fall. Wakeham (294), who is the latest contributor to the subject of the effect of menstruation on the metabolism states that "the considerable daily fluctuations observed in basal metabolism, even when every possible precaution to secure uniformity of conditions is taken, easily masks, in any individual case, smaller variations due to some regular function like menstruation." Wakeham based his conclusions as to the effect

of menstruation on metabolism on a statistical study of 98 observations on 24 women. He found the average of all premenstrual determinations to be 4 per cent higher than the average of all postmenstrual determinations. This is less than the error of the method used.

The reader will probably await further studies of the metabolism in tuberculosis, in view of the differences of the findings of various workers. It should be pointed out that the studies of McCann and Barr were carried out in apparatus in which the chemical measurement of heat was checked by physical measurement, and careful alcohol checks of the apparatus were performed in addition. It is open question as to whether the results obtained with the portable respiration apparatus are sufficiently accurate for experimental work. Outside of the Carnegie Nutrition Laboratory alcohol checks of this apparatus are almost never made. The respiratory quotient is not determined, so that this check is lacking. It is certain that no careful physician would accept the results of a serological laboratory in which all the necessary controls were not made with each Wasserman test.

The mechanism of rising and falling temperature in tuberculosis is shown graphically in a figure taken from the paper by McCann and Barr (190) (fig. 1). There were 5 observations on three individuals. Each pair of columns represent one hour period, the unshaded column showing the heat produced from the body in the same period of time. During the rise of temperature (rectal) heat production exceeded heat loss. At the peak of temperature curve, production and loss of heat were equal. During the fall the loss of heat exceeded the production. In these tuberculous subjects the percentage of heat lost by the vaporization of water increased greatly, amounting to about 38 per cent, whereas Soderstrom and DuBois (278) found it to be very constantly about 24 to 25 per cent in an extensive series of observations. All attempts to observe a patient in the calorimeter during a typical night sweat failed.

Other fevers, which were studied by DuBois and his co-workers, were erysipelas, arthritis, and fever following intravenous injections of typhoid vaccine and of proteose. Erysipelas, which was investigated by Coleman, Barr, and DuBois (65), was found to affect the metabolism in approximately the same way as typhoid fever. One case of arthritis studied by Cecil, Barr and DuBois (60) had an elevation of

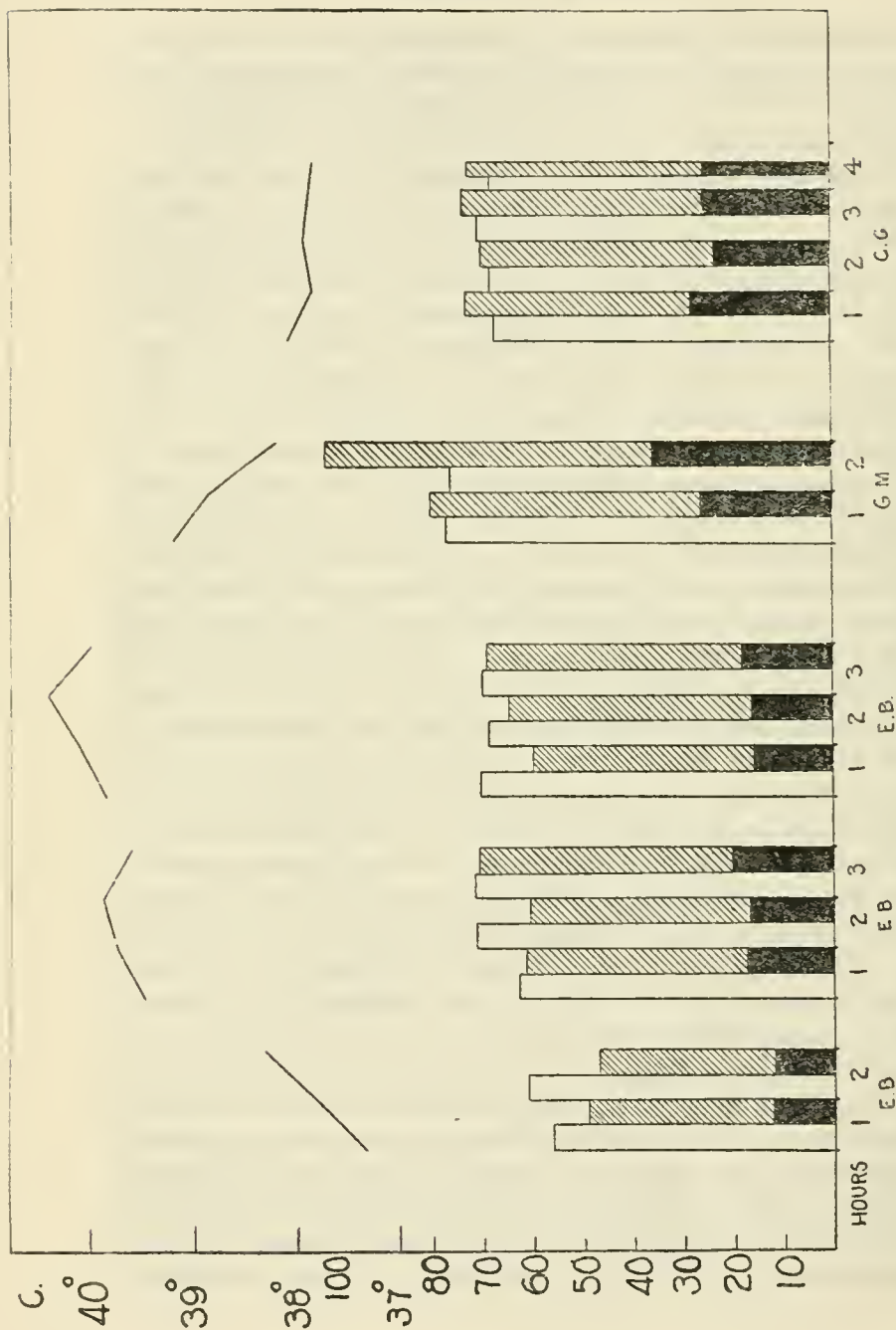


FIG. 1.* RELATIONSHIP OF HEAT PRODUCTION AND HEAT ELIMINATION DURING RISE AND FALL OF RECTAL TEMPERATURE

Five observations on 3 individuals. Lines show rectal temperature in degrees Centigrade. Unshaded columns represent calories per hour produced in the body, determined by the indirect method. Shaded columns represent heat lost from the body by vaporization of water (solid black) and by radiation (hatched). Periods are one hour for each pair of columns. From McCann and Barr (190).

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metabolism which was 26 per cent above the average normal level, when the rectal temperature was 38.4°C. The experiments of Barr, Cecil, and DuBois (22) on the febrile paroxysm following injections of proteose or of typhoid vaccines are most interesting. They found that with the onset of a chill the heat production increased from 75 to 200 per cent, due in part to shivering. At the same time there was almost no increase in heat elimination. The resultant storage of heat elevated the body temperature, which for a short period remained at a level at which heat production and heat elimination were equal, both being 20 to 40 per cent, above the basal level. Following this, as the temperature fell, there was usually a steady decrease in heat production until it reached the normal level. The heat elimination on the other hand increased still farther, thus ridding the body of the stored heat. The method of temperature regulation was almost identical with that of malaria (21).

During the chill these authors noted an increase in respiratory quotient, which they interpreted as being due to a rapid combustion of glycogen stores. It is of interest to compare these findings with those of Lusk (167), who studied the metabolism of normal men during immersion in a bath at 10°C. The shivering produced increased the metabolism as much as 181 per cent. The respiratory quotients rapidly fell to the fasting level in all but one subject. Lusk believed that the quotients fell as the result of the rapid exhaustion of glycogen stores.

From the data of these extensive studies of fever DuBois (78) was able to make the important generalization that the relationship of basal metabolism and body temperature conforms with van't Hoff's law of the effect of temperature on the velocity of chemical reactions. This law may be stated as follows:

With a rise of temperature of 10°C., the velocity of chemical reactions increases between two and three times.

DuBois (78) found that 82 per cent of the observations on fever fell within ± 10 per cent of the average expressed by the temperature coefficient 2.3. In general the observations on fevers in which the protein metabolism was high tended to group themselves about the upper limit (+10 per cent), while those on tuberculosis in which the protein metabolism is relatively low tended to be grouped about the lower limit (-10 per cent).

In accordance with these findings an increase in body temperature of 10°C. should be accompanied by an acceleration of metabolism to 230 per cent of the original value (100 per cent). Ten degrees Centigrade is equal to 18°F. Hence a change of 1 degree Fahrenheit should cause an elevation of $\frac{1.30}{1.8} = 7.2$ per cent (6.5 to 7.9 per cent). In this way one could predict that the basal heat production of a patient with a rectal temperature of 102°F. would be approximately 25 per cent above the afebrile basal level.

It is theoretically possible that an elevation of body temperature might be accomplished by a decrease in heat elimination alone, without an increase in heat production. Leschke (154) reported some experiments purporting to show a reduction of metabolism in what he called "anaphylatoxin fever" in rabbits. Both carbon dioxide production and oxygen absorption of these animals were diminished in spite of a distinct elevation of body temperature. However, in further experiments on dogs direct calorimetric observations were made, which showed a definite increase in heat production in most cases, and in none was there a pronounced decrease. Loening (161) found a fall in temperature and in metabolism during anaphylactic shock. The diminished respiratory exchange of Leschke's rabbits may have been due to a change in the muscular activities of these animals. For experiments on the respiratory metabolism it is hard to find a more unsatisfactory animal than the rabbit, on account of muscular movements and also of the fact that food persists for a long time in the gastrointestinal tract undergoing fermentation with the production of gases which cause errors in the measurement of the respiratory exchange.

Experiments of Freund and Grafe (93) who inoculated poikilothermic dogs with trypanosomes and hog cholera, showed that no increase in metabolism occurred. This would indicate that in febrile infections the increase in metabolism is mediated by the nervous mechanism for temperature regulation and not by direct action of infecting agents or their products on other tissues. Sverzhewski found that animals injected with tetanus toxin showed an increase in metabolism during the latent period before convulsions occurred. It is more than probable that in these experiments the primary effect of the toxin was on the nervous mechanism.

Hibernation. Studies of the metabolism of animals during hibernation are full of matter of the greatest interest to physicians. The occurrence of pseudo-hibernation in man has been recorded by Cushing. Various physiological processes of the hibernating mammal are analogous to certain pathological processes of man and their investigation may cast a great deal of light upon the mechanisms in disease.

A remarkably complete and careful study of the whole physiology of hibernation is that of Raphael DuBois (1896), which includes important results of both direct and indirect calorimetry. This writer reviews the early work of Regnault and Riesel (1849) and of Carl Voit (1878), and of other workers who have studied the respiratory exchange of hibernation. He confirmed the work of Regnault and Riesel who found very low respiratory quotients (0.4 to 0.5) during the winter sleep, though Voit, Mares and others found abnormally high quotients. The observation of high quotients was also verified by DuBois, but only as a transient phenomenon immediately preceding the rise in metabolism which occurs during waking.

The explanation of these low respiratory quotients is very difficult. The favorite hypothesis has been that it is due to the formation of carbohydrate from fat. This view is supported by the observation of Claude Bernard that there is a steady increase of glycogen in the livers of hibernating animals. This increase in glycogen was determined also by DuBois whose figures are given below (80).

Glycogen per 1000 grams of liver
I. Hibernating marmot

Duration of sleep:	grams
4 days.....	6.05
7 days.....	8.88
9 days.....	8.65
10 days.....	16.32

Marmots awake showed only traces of glycogen in their livers. Another interesting observation made by DuBois was the occurrence of a hypoglycemia during the hibernation, and of high blood sugar percentages during and after the awakening.

In the writer's opinion the most reliable information regarding the R.Q. during hibernation is to be found in the work of Henriques (122). He calls attention to the fact that the formation of carbohydrate from fat would yield very low quotients; for example the conversion of olein into carbohydrate would yield a respiratory quotient of only 0.28. Henriques did not observe any animals with a quotient of less than 0.62, and such quotients were noted only when the body temperature was below 10°C.

Another explanation of these observations is that the accumulation of glycogen in the liver of hibernating animals may be due to the failure to oxidize carbohydrate derived from the breakdown of protein. Lusk (170) has shown that similar low respiratory quotients occurring in diabetic subjects may be due to this cause. Similar low quotients, were noted by the writer (189) in an experiment in which meat was given to a normal man at the end of an eight day fast, for which the assumption was made that glucose formed from the protein ingested was stored as glycogen and not immediately burned. The failure to oxidize the carbohydrate moiety of the protein molecule accounts for quotients only as low as 0.62 (170). The finding of lower quotients than this is again reported in the work of Schenk (267) who records respiratory quotients of hibernating hedgehogs between 0.41 and 0.65. There must still remain considerable doubt as to the true nature of the intermediary metabolism during the winter sleep. Whether carbohydrates are formed from fat and protein, or from protein alone the oxidation of carbohydrate is undoubtedly minimal or does not occur at low body temperatures of from 4° to 10°C.

During the waking process a great acceleration of metabolism occurs, heat is stored in the body and gradually the body temperature rises to 36° or 37°C. During this process the respiratory quotients have been found to lie within the usual limits of 0.7 and 1.0. In the experiments of Henriques (122) the quotients ranged between 0.70 and 0.76, in those of DuBois (80) between 0.85 and 0.98, while Schenk records an increase of the respiratory quotient from 0.68 to 0.83.

In the rapid rise of temperature during the awakening from winter sleep the metabolism undergoes changes which are quite analogous to those occurring in fever in man. The initial acceleration of heat production is very marked. The animal at the start is in an almost

lifeless condition, with a rectal temperature of 4°C . and a heat production which is only one-thirtieth to one-fortieth of the normal waking metabolism. Within a period of 5 to 6 hours the temperature rises through 30 to 32°C . This is well shown in an experiment by DuBois (80).

TIME	RECTAL TEMPERATURE	O ₂ CONSUMED PER HOUR
	$^{\circ}\text{C}$.	<i>liters</i>
2:30	11	0.53
4:10	16	1.40
4:40	21	1.80
5:00	26	1.20
5:30	33	0.81
6:10	36	0.35

It is seen that during the early stages of the waking the acceleration of heat production is greatly in excess of that called for by van't Hoff's law, just as it is in man when a rise of temperature is accompanied by a chill.

Basal metabolism in anemias and leukemias. In 1845 Hannover (115) found that there was an increased excretion of carbon dioxide in chlorosis. This result is merely of historical importance. In 1869 Pettenkofer and Voit (241) studied a man with leukemia, whose metabolism was found to be elevated. Bohland (45) in 1893 found a high rate of oxygen consumption by patients with leukemias and in one case of severe anemia from uncinariasis. Two patients with chlorosis did not have an increased metabolism. Unfortunately none of Bohland's patients were "nüchtern," so that the basal metabolism was not measured.

Simple anemia from hemorrhage was studied in dogs by Gürber (111), who found that blood losses up to 3.5 per cent of the body weight (loss of about two-thirds of the red corpuscles) produced no change in metabolism provided that the physical conditions of the circulation were maintained by fluid replacement. Further studies of the same nature were made by Pembrey and Gürber (240). The total metabolism and respiratory quotients underwent no significant alternations following blood losses sufficient to reduce the hemoglobin by one-half.

Grafe (108) studied patients with anemia. The data from five of these can be recalculated for comparison with the Aub and DuBois (10) standards. Of three patients with severe secondary anemia due to gastric ulcer, one had a normal rate of metabolism, and two showed increases of 24 and 25 per cent, respectively, above average normal. Two patients with hemolytic anemia varied from average normal by -11 and $+20$ per cent, respectively.

Eberstadt (82) produced chronic anemias in animals in two ways, by repeated bleeding, and by administration of phenylhydrazin. The effect of these procedures upon the metabolism seemed to depend on the condition of the bone marrow. Usually hyperplasia of the marrow resulted from repeated bleedings, and the metabolism was elevated in the majority of such cases. Phenylhydrazin produced marrow aplasia, and as a rule the metabolism was decreased in such aplastic anemias.

Meyer and DuBois (76) give a table comprising a very extensive compilation of the literature of metabolism in anemias and leukemias. In leukemias of both types the metabolism is shown to be markedly elevated. In chlorosis, secondary anemias, and in pernicious anemia, there is considerable variability in the findings of different investigators. Meyer and DuBois added 5 cases of pernicious anemia in which the variations of metabolism from average normal varied from $+2$ to $+33$ per cent. The finding of normal respiratory quotients indicated that there was no alteration of the normal proportions in which foodstuffs are oxidized.

A very important study of pernicious anemia is that of Tompkins, Brittingham and Drinker (289). Their results showed that the basal metabolism in this disease may be normal in the untreated state. Whatever the original level transfusion invariably diminished it. This suggested to these authors that there is some type of stimulant to the body cells in general in untreated acute cases of pernicious anemia, and that the amount of stimulation is measured by the fall in metabolism resulting from transfusion. Also, they concluded that there are coincident progressive tissue alterations in this disease which tend to reduce metabolism. These alterations were represented by the diminished respiratory exchange of chronic cases and by the low level to which it falls after transfusion.

It is to be noted that the decrease in metabolism following transfusion in pernicious anemia is probably not dependent on the diminution of cardiac and respiratory work which resulted. While it is true that the pulse rate and respiratory activity decreased, the response of the metabolism to transfusion lagged behind that of all other factors by an interval of several days.

That the basal heat production in chronic leukemias may be enormously increased was shown by the work of Grafe (108), Magnus Levy (179), and Kraus (150). It is frequently found to be from 50 to 100 per cent above normal, irrespective of the type of leukemia. Gunderson's (112) results indicate that the basal metabolism in myelogenous leukemia bears a relation particularly to the number of immature leucocytes in the blood stream, irrespective of the total leucocytosis. The highest values are usually found in cases with very high leucocyte counts with many myelocytes or in cases showing high percentages of myeloblasts. Both of these findings indicate great activity of the leucopoietic tissues. In Gunderson's opinion the basal metabolism may be considered an index of this activity. Some doubt may be thrown on this interpretation by the studies of Murphy, Means and Aub (225), who found that treatment with Roentgen rays and radium produced a marked effect in reducing the leucocyte count of a patient with chronic lymphoid leukemia, without producing a corresponding reduction in basal metabolism. In another patient with myelogenous leukemia the metabolism decreased 22 per cent coincident with a decrease in total leucocyte count from 296,000 to 73,000. In these cases, however, the differential counts were not published, so that it is impossible to determine to what extent young and old forms of leucocytes shared in the process of reduction.

The basal metabolism in leukemias is not invariably elevated above normal. In a series of 16 cases studied by Boothby and Sandiford 12.6 per cent were within normal limits (50).

In polycythemia the metabolism may or may not be increased. In Boothby and Sandiford's (50) two cases one showed a positive variation of more than 20 per cent while the other was within normal limits. Fitz (89) quotes unpublished work of Means and Bock, and of Senator, which indicate slight increases in gas exchange in poly-

cythemia. Means and Newburgh (199) reported finding normal basal metabolism in two patients with erythrocytosis due to mitral valvular disease.

Basal metabolism in miscellaneous pathological states. The principal diseases, in which wide variations from the accepted standards of normal basal metabolism may occur, are those of the thyroid gland, febrile diseases, and disorders of the hematopoietic system. The further discussion of this subject will cover a brief review of the results of studies in a wide variety of abnormal conditions in which the basal metabolism has been found to be within or near the normal limits of variation.

Diseases of the circulatory system. Peabody, Meyer and DuBois (233), in their study of patients with cardiac and renal disease, found that those with compensated cardiac lesions showed no increase in basal metabolism. Of twelve patients with *dyspnea*, nine showed a distinct rise in metabolism, and in five of these the increase was from 25 to 50 per cent above the average normal. In this study the agreement of direct and indirect calorimetry was excellent. The finding of normal respiratory quotients indicated that there was no disturbance of the intermediary metabolism. A further study was made by Peabody, Wentworth and Barker (235). The patients were divided into two groups according to the degree of reduction of the vital capacity of the lungs. The first group comprising those whose vital capacity was over 60 per cent of the normal, showed an average variation of +2.5 per cent and a range of variation of -7 to +19 per cent of average normal. The second group comprising patients with a vital capacity of less than 60 per cent of normal, showed an average variation of +12.8 per cent and a range of -6 to +40 per cent of average normal metabolism.

During the war Peabody, Wearn and Tompkins (234) made a study of the basal metabolism of soldiers with "irritable heart." The majority of these patients proved to have a metabolism within normal limits, which led the investigators to conclude that hyperthyroidism did not play a significant rôle in the production of the symptom complex. Boothby and Sandiford (50) report essentially the same results in 99 cases of "cardiac neurosis."

"*Essential hypertension*" was also studied by Boothby and Sandiford (50). Of 170 patients, so diagnosed, about 90 per cent proved to have a rate of metabolism within ± 15 per cent of normal, the remainder being moderately increased above normal.

Nephritis was investigated by Aub and DuBois (12). The most significant alterations in the heat production per unit of body surface were noted in patients with oedema. In two such cases the basal metabolism was 27 and 40 per cent below average normal. One patient in uremia was found to have a normal rate of heat production. Of those patients with nephritis, hypertension and associated cardiovascular changes the basal metabolism was normal.

In *Gout* the basal metabolism lies within normal limits according to the studies of Wentworth and McClure (296). No changes in the intermediary metabolism could be detected by the respiratory quotient, which lay within normal limits. Like figures were obtained also in one case studied by Cecil, Barr and DuBois (60). In the early literature of Magnus Levy (181) similar results were recorded.

Cecil, Barr, and DuBois (60) also observed patients with various forms of *arthritis*, acute, subacute, and arthritis deformans. There was nothing to indicate abnormality of heat production in afebrile periods. Previously Pemberton and Tompkins had reported observations made upon 29 soldiers with arthritis (239). Of these all but seven were within normal limits, and only two had a basal metabolism more than 15 per cent below average normal. Of 69 cases reported by Boothby and Sandiford 64 were within the limits of ± 15 per cent, 4 were slightly lower and 1 slightly higher (50).

In *obesity* the basal metabolism per unit of body surface is generally normal according to Means (200, 202), who has made excellent studies of this subject. When the basal gas exchange was below normal Means found that there was generally some evidence of disturbed internal secretion. More recently in the larger series of Boothby and Sandiford 95 per cent were found to have a basal metabolic rate within 15 per cent of the average normal (50).

Six cases of Mongolian idiocy were studied by Fleming (90), who found a normal basal metabolism in all according to Benedict and Talbot's standards (38). The surprising fact was noted that the administration of thyroid extract did not increase the heat production

of these children as it did in the case of cretins, whose metabolism was subnormal.

Scattered throughout the literature are observations of the metabolism in various nervous and mental diseases. One of the earliest was the study by Magnus Levy of a patient with *paralysis agitans* (184). The continuous tremor of this disease considerably increases the basal heat production. Magnus Levy observed that the administration of hyoscin diminished the metabolism, presumably because of its effect on the tremor. Boothby and Sandiford (50) record the finding of a normal metabolic rate in all of their cases of *tabes dorsalis*, *migraine*, and nearly all of their patients with "chronic nervous exhaustion" and "neurasthenia." In one-fifth of their cases of epidemic *encephalitis* the metabolism was increased. To what extent this was due to fever, or to involuntary movements from extra-pyramidal motor disturbances cannot be determined from the data published.

In *stuporous* conditions and *catatonia* Grafe (104) reported finding a reduced metabolism. It is hard to judge of the value of his conclusions. He used as the normal standard for heat production 800 calories *per diem* per square meter (Meeh). The height of his patients was not recorded. Obviously the heat production of some patients was quite low. From the meagre data given it is impossible to determine what rôle inanition played in the production of the low metabolism.

Dementia praecox and *manic depressive insanity* were investigated by Gibbs and Lemcke (99). In the dementia group the most marked variations from normal were observed, generally below normal. It was not thought that inanition could account for the low metabolism, nor did the patients show sufficient evidence of thyroid or pituitary disorders to explain the findings satisfactorily. Several of the dementia praecox patients did show evidence of disturbed growth, including incomplete sexual maturity.

Liver extirpation. Mann at the Mayo Clinic has called attention to the development of hypoglycemia in dogs following extirpation of the liver, which had been described by Minkowski, (212) in 1886. As a result of study of the respiratory exchange of these dogs before and after the operation Mann (185) found that carbohydrate was

being utilized after hepatectomy and the injection of glucose, suggesting that the hypoglycemia following this operation is due to utilization of carbohydrate. This revives an old question concerning the significance of the R.Q. after liver extirpation.

Grafe and Deneke (107) noted an elevation of the respiratory quotients after liver extirpation, which they attributed to a diminished alkalescence of the blood. Murlin, Edelman, and Kramer (221) found that clamping the abdominal vessels gave rise to an increase in respiratory quotients, but the evidence indicated that this elevation of R.Q. was due to pumping out CO₂ by over-ventilation since the CO₂ content of the arterial blood was found to have decreased. Without careful study of the blood gases and of the acid base equilibrium it would be unsafe to accept the view that the rise in R.Q. really indicates increased carbohydrate utilization.

Fischler and Grafe (105) noted a marked fall in heat production following ligation of the hepatic artery two to three weeks after establishing an Eck fistula. In the earlier extirpation experiments of these authors (88) the heat production decreased from 30 to 70 per cent. This result was probably due to traumatic shock (14).

Diseases of the liver. Aub and Means (15) reported metabolism determinations of a number of patients with various diseases of the liver and bile passages. These included cirrhosis, neoplasms, gall stones, and catarrhal jaundice. The metabolism in all of these fell within the normal limits of the DuBois surface area formula.

Traumatic shock was found by Aub (14) to produce a marked diminution of metabolism, depending on the severity of the shock. It may fall to as much as 70 per cent below the normal level. A similar decrease of the basal metabolism may be rapidly accomplished by interference with the circulation or by increasing the intrapericardial pressure. The effects of hemorrhage were not constant, occasionally a temporary lowering of metabolism was noted. The recovery from shock was accompanied by a return of heat production to normal.

Diseases of ductless glands other than thyroid. *Adrenals.* The metabolism data from a number of cases of Addison's disease were collected by Aub, Forman, and Bright (17). These have all shown a moderately reduced basal metabolism. Boothby and Sandiford (50)

studied 13 cases, in two of which the metabolism was more than 20 per cent below average normal. Loeffler (160) who was unfamiliar with modern work in normal standards, recorded data in two cases of Addison's disease which the writer has recalculated. These patients showed less than 5 per cent deviation from the Aub and DuBois (10) average normal. Muirhead (214), reporting his own case, states that his basal metabolism was found at the Mayo Clinic to be 30 per cent below average normal.

Experimental adrenalectomy was performed on cats by Aub, Forman and Bright (17). At the end of 48 hours after operation it was found that the metabolism was 25 per cent below normal. No change in intermediary metabolism was detected. Control experiments on fasting cats and on cats, which had been subjected to other operative procedures, showed a decrease in heat production of less than one half the magnitude of that following adrenalectomy. When one adrenal only was removed there was only a temporary decrease in gas exchange.

Marine and Baumann (187) performed experiments on rabbits in which either adrenalectomy was performed or else the adrenal was injured by freezing. These authors reported that such procedures generally increased the metabolism as measured by the gas exchange. An examination of their protocols reveals great irregularities in metabolism from day to day. The respiratory quotients were very irregular. Some of the experiments show a distinct decrease in metabolism almost as frequently as a rise. No method of recording activity of the animals was used. In view of these facts these authors do not seem to be justified in the conclusions which they have drawn from the data presented.

These experiments were repeated on cats by Scott (269). This writer found that "sufficient" non-fatal injury to the suprarenal cortex by freezing or ligation caused a significant increase of metabolism. The experimental protocols of this work appear to be more satisfactory than those of Marine and Baumann. The cat is undoubtedly much superior to the rabbit for determination of respiratory exchange. Satisfactory records of movements of the animal and checks of the apparatus were not recorded.

Marine and Baumann (188) and Scott (269) have endeavored to show that there is an interrelationship between thyroid and adrenals, indicated by the finding of an increased heat production and thyroid hyperplasia in animals with suprarenal insufficiency. Marine and Lenhart (186) found that adrenalin injected into normal and thyroidectomized animals produced relatively the same increase of oxygen consumption in each. In the thyroidectomized animals the increase was delayed and of shorter duration than in the normals.

Grave doubt as to the existence of an interrelationship between thyroid and adrenals is cast by the work of Aub, Bright and Uridil (16). These investigators found that the adrenal glands were not essential to the maintenance of a high metabolic rate induced by administration of thyroxin. Added to this is the evidence furnished by Sandiford (263), who found that the intensity of the reaction to injection of epinephrin was independent of the state of thyroid function. Her 46 subjects included some patients with both hyperthyroidism and some with hypothyroidism.

The effects of complete adrenalectomy have been studied by Gradinescu (101) and by Aub, Bright and Forman (17). These authors both record a pronounced decrease in heat production. Gradinescu did not record the blood pressure and temperature of his animals, so that it is impossible to determine to what extent traumatic shock was responsible for the diminished metabolism. However, Aub and his coworkers found that even when blood pressure and temperature were kept normal the metabolism decreased markedly about 24 hours after operation. This continued until the terminal collapse, at a level about 25 per cent below the previous normal.

The decrease in metabolism following thyroidectomy occurs slowly. The increase produced by administering thyroid extract or thyroxin likewise occurs slowly, requiring several days for the production of the maximum effects. The diminution produced by adrenalectomy occurs quickly. Injection of adrenalin produces an almost instantaneous increase in heat production. In summing up the evidence Aub and Taylor conclude that both thyroid and adrenals affect the metabolism markedly but independently, that the thyroid acts slowly and continuously, the adrenal quickly and transiently.

The pituitary gland. The studies of metabolism following hypophysectomy indicate that a diminution occurs. This conclusion is by no means certain. The careful work of Benedict and Homans (29), in which there was found to be a decrease in CO_2 production per kilogram of body weight following hypophysectomy, has been criticized by Aub and Taylor on the ground that other factors may have been operating to produce the decrease. For instance Lusk (171) showed that mere retention of animals in cages may cause a decrease in rate of metabolism.

In certain cases of obesity, in which the heat production per unit of body surface was below normal, Means (200) found evidences of disordered function of the pituitary. In 58 patients, with the diagnosis of hypopituitarism, Boothby and Sandiford (50) found 55 per cent to be within normal limits, 6.8 per cent were more than 15 per cent above average normal, and 38 per cent were more than 15 below average normal. Of 30 cases of acromegaly studied by the same authors about one-third were above the upper limit of normal (+15 per cent). One case was more than 20 per cent below average normal.

It would be unsafe to conclude that these findings indicate that the normal secretory products of the pituitary body influence the rate of metabolism. Diseases of the pituitary body are frequently associated with disorders of other ductless glands. Regarding the evidence of the effects produced by the ingestion or injection of pituitary extracts more will be said under the heading of the effects of drugs. It is very difficult to prove that extracts derived from glands of internal secretion are identical with the secretory products of these glands during life. For this reason conclusions regarding the function of a gland should be drawn with great caution from experiments on the effects of administration of an extract of the gland. Boothby states that "there is, however, as yet little evidence that the secretion of any part of the pituitary gland is concerned with the normal rate of cellular combustion in the sense that it acts as a calorogenetic agent."

Diabetes insipidus. Five cases of this disorder were studied by Snell, Ford and Rowntree (277). Two of the five patients showed abnormally high rates of metabolism on initial observations. These patients later showed normal metabolism after treatment with

pituitrin. The metabolism of the other three patients was not affected by pituitrin, though in all five cases it was successfully used as an anti-diuretic agent.

Gonads. In 1895 Caratulla and Tarulli (55) measured the respiratory exchange before and after castration of one female dog and two mice, using a small Pettenkoffer Voit Apparatus. The dog showed a gain in weight of 17 per cent and a decrease in CO_2 production of 34 per cent following castration. Similar results were obtained with the mice.

Loewy and Richter (163) castrated a female dog. After 10 to 14 weeks the weight of the dog had increased 10 per cent, while the oxygen consumption per kilogram had decreased about 20 per cent. A male dog was castrated, but in this case the dog lost weight in the same proportion as the decrease in metabolism.

Luethje (164) studied the respiratory exchange of a castrated female dog using a normal bitch as a control. Both were fed on the same diet, but observations were not made under the conditions of basal metabolism. His results are of no value for our present purpose.

Zuntz (303) gave the results of metabolism experiments on four women who had been castrated. Only one subject showed a clear decrease. The data are not worth much as the patients took food and were restless with pain.

Murlin and Bailey (217), (20) studied two dogs after oöphorectomy. Their results upheld the work of Loewy and Richter. Removal of the ovaries of their dogs was followed by an increase in weight in both and a lowering of the metabolism by 6 to 14 per cent in one and by 12 to 17 per cent in the other. No marked effect on the heat production was noted until 18 to 21 days after operation. Murlin and Bailey did not believe that the lowering of metabolism was a direct result of oöphorectomy, and that the indications did not point decisively to the loss of a specific stimulus from the ovary affecting the oxidative processes of the cells.

Spleen. Danoff (67) found a marked increase in metabolism of white rats after splenectomy. The animals died soon after operation. No data are given as to the presence or absence of infection or fever. Danoff worked in Asher's laboratory. Two years later Asher and Koda (8) reported that removal of the spleen from a dog did not increase the basal metabolism. Korentchewsky (147) studied the

effect of removal of the spleen of three dogs. He was unable to find any effect upon the respiratory metabolism. Tompkins, Brittingham and Drinker (289) reported that the metabolism of two patients with splenic anemia was lowered following splenectomy. In summary it may be said that the spleen does not appear to have any direct effect upon the rate of metabolism.

Parathyroid. The writer has been unable to find satisfactory experimental evidence concerning the effect of parathyroid extirpation on the heat production. In some cases the result is obscured by the coincidence of thyroid insufficiency due to simultaneous thyroidectomy. In other cases the evidence of complete removal of parathyroid tissue is lacking.

Tetany, resulting from excessive pulmonary ventilation, is accompanied by an increase in oxygen consumption according to the admirable experiments of Grant (110). In order to show that this increase of metabolism was not due to the increased muscular work of breathing, the experiments were controlled by having the subject breathe the same amount of air containing 5 per cent CO_2 . When the excessive loss of CO_2 during forced breathing was prevented, the oxygen consumption in the control periods was from 10 to 20 per cent less than in the periods with hyperpnea with air alone.

Loeffler (160) reported a case of parathyroid tetany following thyroidectomy, in which the metabolism was 18 per cent below the average normal of Aub and DuBois (10).

Myasthenia gravis. Loeffler (160) reported observations of a woman, thirty years old, whose metabolism on recalculation is found to be 10 per cent below the average normal.

Pancreas. Benedict and Joslin in 1910 made a study of the total metabolism of patients with *diabetes mellitus*. They concluded that the average heat production of their patients was 15 per cent above that of their normal controls, the comparison being made on the basis of body weight. Lusk (168) criticized their conclusions, and recalculated their results, with the conclusion that the average heat production was only 5 per cent above normal in diabetes. At this time the subject of normal standards was in an unsatisfactory state.

The writer has recalculated the metabolism of 19 subjects of Benedict and Joslin (30), and compared the results with the average normal

standards of Aub and DuBois (10). The metabolism of these patients varied from 27 per cent below to 32 per cent above average normal, with an average deviation of 4 per cent.

From the writer's own series of diabetic subjects 51 observations on 26 patients showed deviations from average normal ranging from -38 per cent to +12 per cent, with an average deviation of -8.25 per cent.

When repeated observations are made on patients who are undergoing treatment, it is noteworthy that improvement in the patient's condition or carbohydrate utilization is almost invariably accompanied by a slight reduction in the basal metabolism. This fact was pointed out by Wilder, Boothby and Beeler (297). It is amply confirmed by the writer's own observations (194), (195). These authors (297) state that the metabolism in diabetes is never above the average normal of DuBois, unless there is a complicating hyperthyroidism, or unless infection is present, or the patient has been given an excessive diet, especially if protein be in excess. They state that the finding of a normal basal metabolism in an emaciated patient indicates that the patient is receiving too much food, or else is in a critical stage of starvation in which an excessive loss of body protein is taking place.

The foregoing conclusions in regard to the basal metabolism in diabetes are borne out in the latest publication of Joslin's (138) results. In this report Joslin makes an extensive analysis of all factors which may enter into the interpretation of the results.

It is quite apparent that the determination of the basal metabolism of diabetic patients has some value from a prognostic standpoint, and as an indication of progress of therapy. The most important use of the determination is that of establishing the minimal food requirements of the patient. This has proved of great value in the writer's practice in which the usual procedure is to give the patient a diet furnishing only enough energy to cover the minimal requirement (basal metabolism plus 10 per cent) until "desugarization" was accomplished (195). With the newer knowledge of antiketogenesis it is possible to prescribe diets in such a way that the energy requirement will be filled by a mixture of foodstuffs containing only a minimum of antiketogenic substances (glucose or substances capable of conversion into glucose (114)).

i. The effects of drugs on the basal metabolism

Thyroid extracts. Thyroxin. There have been many calorimetric studies which showed that the administration of thyroid extract increased the heat production of normal subjects and of those with hypothyroidism. The earliest of these studies was made by Magnus Levy (178). As typical examples of the method of accurate calorimetric control of the dosage of thyroid preparations in hypothyroidism the reader is referred to the work of DuBois (75), Means (205), Talbot and Moriarty (285), and of Fleming (90). It is interesting to note that Fleming (90) was unable to increase the metabolism of Mongolian idiots by administration of thyroid extracts, which were effective in cases of cretinism. The action of thyroid extracts is not predictable, owing to the varying potency of different preparations. The advent of the pure thyroxin, which was isolated by Kendall, marked a great advance in the therapy of hypothyroidism, because with this drug the effect upon the heat production may be predicted with a reasonable degree of certainty. Under the heading of the basal metabolism in thyroid disease a table is given, which was taken from the work of Boothby and of Plummer and Boothby, in which one may find the necessary data for computing the dosage of thyroxin in terms of its metabolic effect.

The importance of controlling the administration of thyroxin by metabolism determinations cannot be over-emphasized. It is true that under the influence of the thyroid variations in the level of basal metabolism are accompanied by proportionate variations in the *basal* pulse rate in the vast majority of cases, as is shown by the work of Sturgis and Tompkins. However, cases are continually encountered in which some abnormality of the cardiac mechanism prevents the usual response in heart rate to change in metabolic rate. Such cases are well illustrated by the report of Aub and Stern (13), whose patient had a complete auriculo-ventricular dissociation. The ventricular rate did not increase in response to over-dosage with thyroid extract, sufficient to elevate the basal metabolism to a level 47 per cent above normal and to increase the auricular rate to 120.

The calorogenetic action of thyroxin is probably due to a direct stimulation of the cells to a higher rate of oxidation. As a result of

their experiments, Aub, Bright and Uridil (16) concluded that it could not be explained by increased muscular activity, muscular fibrillation or increased tonus. These authors also found that the presence of the adrenal glands was not essential to the maintenance of a high metabolism by thyroxin.

Epinephrin. An enormous number of experiments have been made concerning the effect of epinephrin on the respiratory exchange. With one exception all investigators have noted an increase in metabolism following the administration of epinephrin to normal men and animals. Hari (117), alone, reported a diminished consumption of oxygen. This may have been due to the fact that Hari worked with curarized animals. Reliable observations, in which oxygen consumption and heat production have been increased by injection of epinephrin, are recorded in the work of Lusk and Riche (176), Tompkins, Sturgis, and Wearn (290), Sandiford (263), and others. The effects produced in normal subjects by the subcutaneous injection of 0.5 cc. of a 1:1000 solution of epinephrin have been well standardized. These effects are well illustrated in the graphical record of experiments on four normal men by Lyman, Nicholls, and McCann (177) in figure 2. The heat production, calculated from the oxygen consumption, increases markedly within 10 minutes after injection of epinephrin, reaching a maximum within about 30 minutes. The respiratory quotient increases in nearly all cases within the first 10 to 20 minutes, thereafter decreasing until it falls below the initial level in from 30 to 60 minutes. This increase in respiratory quotient has usually been supposed to indicate an increased combustion of carbohydrate, coincident with the rapid glycogenolysis induced by epinephrin. The rapid mobilization of carbohydrate from the stores in the liver results in hyperglycemia in normal individuals. The elevation of the blood sugar curve does not reach a maximum until from 30 to 60 minutes, at which time the respiratory-quotient is decreasing. This fact was noted by Bornstein and Müller (52), who also called attention to the fact that, in the initial period in which the R.Q. is increasing, the alveolar CO_2 tension decreases markedly. This observation was also confirmed by Lyman, Nicholls and McCann (177), who found marked reduction in alveolar CO_2 tension, great increase in minute volume respired, in tidal air, in the calculated dead

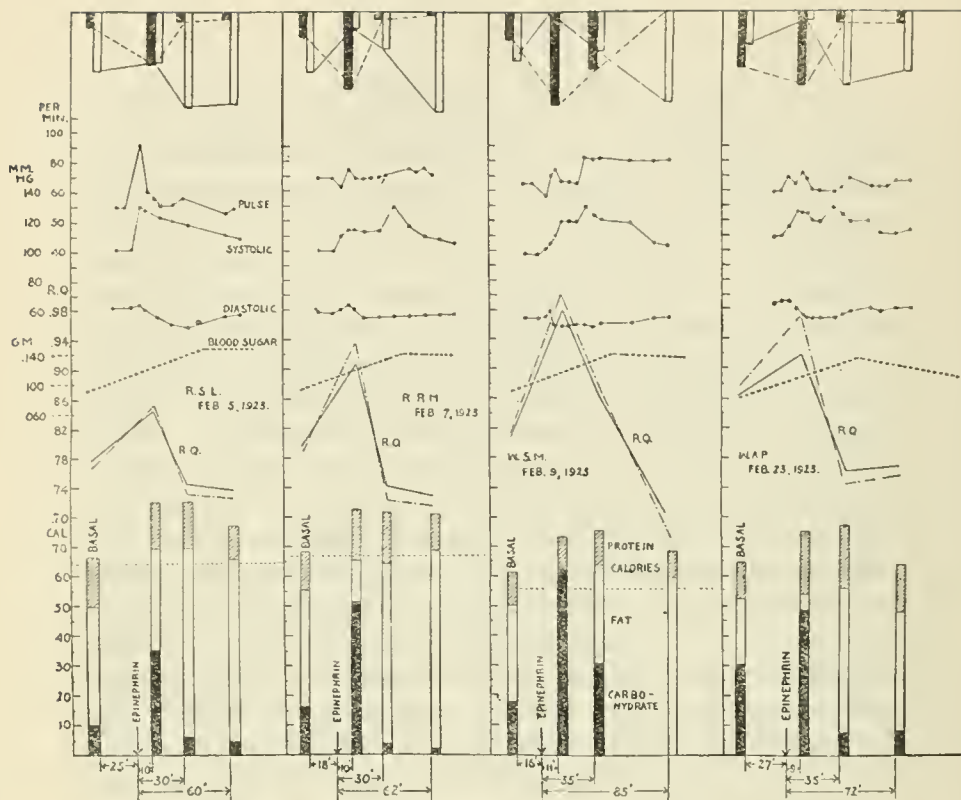


FIG. 2.* EFFECTS OF 0.5 CC. EPINEPHRIN IN NORMAL SUBJECTS (177)

The abscissa units represent time in minutes, labeled arrows indicating the injections of epinephrin or insulin.

Items on the ordinate axis from the bottom up are represented as follows:

1. Calories per hour in each successive respiratory period as vertical bars. A horizontal broken line gives the normal standard of calories per hour for the sex and age and surface area of the subject in each experiment, according to the method of Aub and DuBois.

When the bars are subdivided, black represents calories derived from carbohydrate and white calories from fat metabolism, as calculated from the non-protein R.Q. The cross-hatched portion denotes calories derived from protein as calculated from the urinary nitrogen.

2. Total R.Q. at each respiratory period as solid lines. Broken lines accompanying the graph of total R.Q. represent the non-protein R.Q., when plotted.

3. Broken line as blood sugar in grams of sugar per 100 cc. of blood.

4. Diastolic and systolic blood pressure as solid lines on which the time of successive readings is indicated by dots.

5. Pulse rate as solid lines with dots indicating the time of consecutive determinations.

* This figure is reproduced through the courtesy of the Journal of Experimental Pharmacology and Therapeutics.

space, and in the effective alveolar ventilation. These observations all point to a new explanation of the increased R.Q. after epinephrin; namely, that it is due to stimulation of the respiratory mechanism to over-ventilation with consequent washing out of carbon dioxide from the blood and tissues. The question must still be considered unsettled as to whether a true increase in carbohydrate oxidation follows the administration of epinephrin or whether the change in respiratory quotient is the result of changes in the mechanism of breathing induced by the drug.

Additional light is thrown on this subject by the studies made by Lyman, Nicholls and McCann (177) of the effect of epinephrin on diabetic subjects. In diabetes mellitus the ability of the organism to store carbohydrate as glycogen is usually found to be impaired. Also, as a rule, the increase in blood sugar concentration produced by epinephrin is usually much less in diabetes than in health. The respiratory quotients of diabetic patients were sometimes not increased at all, and in other cases were increased much less than in normal subjects. Indirectly this supports the view that the respiratory quotient after epinephrin may depend, in part at least, on the extent of carbohydrate oxidation.

The effect of epinephrin on the heat production of diabetics varied. Half of the patients showed an increase of 6 to 18 per cent, which was less than the normal increase. The other half showed an increase in metabolism of 29 to 43 per cent above the basal level. The average increase in normal subjects lay between 17 and 33 per cent (177).

Sandiford (263) raised the question as to whether the increased metabolism following epinephrin might not be due to the carbohydrate plethora. However, Boothby and Sandiford (48) found that the ingestion of enough glucose to double the blood sugar percentage increased the heat production by 9 per cent; whereas, a dose of epinephrin which produced an elevation of blood sugar of only 37 per cent increased the metabolism 20 per cent. This would indicate that epinephrin possesses a specific calorogenetic action.

Sandiford (263) called attention to the fact that the metabolic effects of epinephrin injections were independent of the degree of hyper- or hypothyroidism. Marine and Lenhart (186) found that the effect in thyroidectomized rabbits was generally somewhat delayed and of shorter duration than in normal animals.

Insulin. In the early studies of the physiological effects of insulin Banting and his collaborators reported that the administration of insulin caused an elevation of the respiratory quotient of diabetic subjects. This indicates that insulin increases the oxidation of carbohydrate.

Lyman, Nicholls and McCann (177) observed the respiratory exchange of normal and diabetic subjects before and after injection of insulin intravenously. In all experiments on normal subjects there was a marked increase in R.Q., reaching a maximum about 30 minutes after intravenous injection of $3\frac{1}{2}$ units of insulin. The rise in R.Q. is not so prompt as in the case with epinephrin, and may not occur until a considerable decrease in blood sugar has occurred. The heat production was increased in all cases, reaching a maximum of 2.5 to 17.7 per cent above the basal level in from 10 to 60 minutes. At the end of $1\frac{1}{2}$ to $2\frac{1}{2}$ hours after injection the heat production was again at or below the basal value. These subjects all received maximal or nearly maximal intravenous doses. The degree of increase in metabolism of diabetic subjects was observed to be approximately the same as with the normal controls, ranging from 2.9 to 19.6 per cent, with an average value of 11.6 per cent above the basal level. It was found that epinephrin and insulin were antagonistic, to a certain extent. When epinephrin was given 22 minutes after insulin the rise in heat production was much less than when epinephrin was given alone, but more than when insulin was given alone.

Murlin (224) and his coworkers have reported elevation of the R.Q. after administration of aqueous extracts of pancreas, and have called attention to similar findings by Murlin and Kramer as early as 1916 (223).

Pituitary extracts. A dried preparation of the anterior lobe was injected into two dogs by Benedict and Homans (29), with the result that body temperature and carbon dioxide elimination were increased in both instances. This result may not be specific, and may be entirely due to the introduction of foreign protein substances, as in the case of proteose and typhoid vaccine studied by Barr and Cecil and DuBois (22).

Bernstein (40) found that the metabolism was depressed following the administration of an extract of the anterior lobe of the pituitary.

When extracts of the posterior lobe were given the metabolism increased from 19–25 per cent. Snell, Ford, and Rowntree (277) found that injections of pituitrin (posterior lobe) did not increase the heat production of patients with diabetes insipidus, a result which the author was able to confirm in one instance. Changes in metabolism found by McKinley (197) following posterior lobe injections were quite small.

The antipyretics. *Sodium salicylate* was given by Denis and Means (68) in doses up to 6.6 grams *per diem* without marked effect upon the heat production. In one case with symptoms of salicylate intoxication the metabolism was elevated 18 per cent above the normal basal level.

Barbour and Devennis (19) found that *acetyl salicylic acid*, given to normal men in 1 gram doses produced a slight increase in heat production, without significant alteration in heat elimination, body temperature, or respiratory quotients. However, Barbour found that when *acetyl salicylic acid* was given in 1 gram doses to patients with fever, the heat elimination was increased by 38 per cent, and, as the body temperature fell the heat production diminished.

Berrar (42) found that the administration of large doses of *aloin* produced fever, and in consequence an elevation of metabolism was found. Some of the experimental subjects of Carpenter and Benedict (57) developed fever from breathing mercury vapor in the respiration apparatus, an increase in metabolism being noted.

The administration of 1 mgm. of *atropin* was found by Higgins and Means (125) to increase the metabolism. In a previous publication Edsall and Means (83) found that $\frac{1}{25}$ grain of atropin increased the oxygen consumption about 15 per cent, and that $\frac{1}{50}$ grain—increased it about 8 per cent.

Magnus Levy 184 found that the administration of *hyoscin* to a patient with paralysis agitans caused a diminution in respiratory exchange. This effect is undoubtedly due to the effect of the drug in diminishing the tremor and muscular rigidity of this disease.

Jackson (134) studied the oxygen consumption before and after the administration of various drugs, using a recording spirometer, with oxygen in a closed circuit in which CO_2 was continually absorbed. Volume changes due to bronchial constriction produced temporary

changes in the graphic record of oxygen consumption. *Arecoline* produced a bronchial constriction, without marked effect on the rate of oxygen absorption. *Pilocarpine* also produced bronchial constriction, less marked than that due to arecoline. Subsequent to this pilocarpine seemed definitely to increase the metabolism. The experiments of Börnstein and Müller show an increase in the oxygen consumption after pilocarpine though it should be noted that the animals had been vomiting and retching. This drug was found by them to cause a slow steady rise in blood sugar and coincidentally a slow rise in the value of the respiratory quotient. Since the alveolar CO₂ tension was not decreased the rising R.Q's were taken as evidence of increased oxidation of carbohydrate. Kelemen (140) found that pilocarpine in doses of 0.001 gram per kilogram of body weight produced an increase in metabolism of about 10 per cent, with a slight rise in respiratory quotient.

Caffeine. In 1859 Edward Smith (275) found that after taking 50 to 100 grains of tea leaves or one-half cup of strong coffee a rise of carbon dioxide output was invariably observed in several normal subjects. The rise was usually from 15 to 30 per cent. In 1890 E. T. Reichert (251) published experiments on the effect of caffeine on dogs studied by direct calorimetry. Very large doses were used. A dose of 35 mgm. per kilo of body weight produced a rise in heat production of 38.6 per cent; 70 mgm. per kilo, a rise of 43.6 per cent, and 100 mgm. per kilo a rise of about 70 per cent. Edsall and Means (83) found that with man 5 grains of caffeine sodium salicylate increased the oxygen absorption per minute from a basal value of 275 cc. to 316 cc. an increase of 15 per cent. Higgins and Means (125) also found that caffeine increased the respiratory exchange. Using the respiration calorimeter of the Russell Sage Institute of Pathology, Means, Aub and DuBois (204) found that when caffeine was given in doses of 8.6 mgm. per kilo of body weight to 4 normal subjects the heat production was increased from 7 to 23 per cent above the basal value. No changes in the intermediary metabolism affected the respiratory quotient.

Higgins and Means (125) found that the usual therapeutic doses of *strychnine* did not raise the rate of metabolism. With doses as large as $\frac{1}{15}$ grain a slight increase was noted by Edsall and Means

(83). No studies of the effect of convulsant doses have been made but these would undoubtedly show a pronounced increase in heat production. It is of interest in this connection to note that Brunner (54) found an elevation of metabolism during the latent period after injection of *tetanus toxin* in large doses.

Camphor was found by Higgins and Means (125) to cause an increase in metabolism. In the previous studies of Edsall and Means (83) the results with camphor were inconclusive.

Opium alkaloids. In 1898 Dreser (70) found that morphin, heroin and codeine decreased the metabolism. His experiments were performed on animals, and as no quantitative record of the activity of the animals was kept, one may pertinently raise the question as to whether or not the decrease was due to a diminution of muscular movements. Higgins and Means (125) found that morphin and heroin in therapeutic doses produced practically no effect on the metabolism. Chanutin and Lusk (61) obtained variable results. After receiving 15 mgm. of morphin per kilo one dog showed an average reduction in heat production of 6.2 per cent (23 per cent at the maximum). With the second dog in spite of the increased heat production there was excessive heat loss so that the body temperature fell, the pathways for the distribution of heat loss being unaltered.

Jackson (134) found that very small doses of *sodium cyanide* slightly accelerated oxygen consumption, but that larger doses retarded it. In three out of four patients with hyperthyroidism, Snell, Ford and Rowntree (277) found that the basal metabolism was higher while sodium cyanide was being given in doses of $\frac{1}{20}$ grain twice daily.

Urethane was found by Aub, Bright and Uridil (16) to increase the metabolism. This effect was noted with both normal and thyrotoxic animals. An average increase of 15 per cent was observed when urethane was given to four normal cats, and an average increase of 13 per cent in ten thyrotoxic cats. Aub, Bright and Forman (17) found evidence suggesting that the flow of adrenal secretion was greater in animals under the influence of urethane. In his monograph on the respiratory exchange Krogh (151a) reviews the literature in regard to the effects of narcotics of this class on the oxidation of unicellular organisms, finding that the oxidations are diminished. In view of this fact the effects of urethane on mammals is rather surprising.

j. Summary

The facts concerning the basal metabolism, which have been reviewed, show that there are many conditions, both normal and pathological, in which variations from the average normal standards are encountered. The metabolism of normal adults, referred to surface area, varies over a range of 30 per cent (± 15 per cent of average normal). It is probable that the metabolism of children varies over a still wider range. The pathological states in which the greatest deviations from average normal are found are those of the thyroid gland, fevers, and diseases of the hematopoietic system. Smaller variations have been observed in a great number of diseases. These facts should make it clear that the diagnostic value of metabolism determinations is no greater than that of a total leucocyte count.

If the basal metabolism is determined by the most accurate technique, under rigidly controlled conditions, a deviation from the average normal can be properly interpreted only in the light of the most thorough and painstaking clinical study of the case. In the last analysis a diagnosis must rest upon the clinical observations.

As an unbiased objective method of evaluating the results of therapeutic measures in thyroid disease, especially as a means of controlling the treatment of hypothyroidism basal metabolism determinations are invaluable.

The basal metabolism represents only a portion, often a small portion, of the total daily energy transformations in health and disease. For a more complete understanding of the food requirements it is necessary to know the effect of food ingestion and the energy expenditure during muscular work. When the total metabolism is considered in this way many important correlations can be made between it and the functions of heart, lungs and blood, which will greatly elucidate the symptomatology of certain diseases.

PART II. THE EFFECTS OF INGESTION OF FOOD ON THE METABOLISM
IN DISEASE

a. The specific dynamic action of foodstuffs

An excellent review of the physiological aspects of the specific dynamic action of the various food factors has recently been made by

Lusk (175), who is one of the principal contributors in this field. Lusk's conceptions of the phenomena following the ingestion of food-stuffs is somewhat as follows:

Under post-absorptive conditions the tissue cells, bathed in a medium in which the concentrations of nutrient particles are fairly constant, maintain a constant level of minimal metabolism, known as the basal metabolism. During digestion and absorption glucose, fat and certain amino-acids increase in concentration in the nourishing plasma surrounding the tissue cells, and under these circumstances the production of heat by an animal increases. The increase of heat production under these influences differs with the different kinds of foodstuffs administered.

The ingestion of carbohydrate induces a carbohydrate plethora in the nutritive environment of the cells to which the cells react with an increased metabolism, except when the extra glucose is being continually depleted under the influence of work, or is reduced, as in fasting, when the blood stream is also under the regulatory control of the liver.

In like manner Lusk (175) explains the specific dynamic action of fat as a "metabolism of fat plethora, due to the utilization of fat by special fat receptive cellular affinities."

"The specific dynamic action of protein consists in a specific chemical stimulus of the cellular protoplasm, which is independent of the oxidation of the cellular material through which the stimulus is applied. It may be termed the metabolism of amino acid stimulation" (Lusk). Glycocoll and alanine are the two amino acids having the greatest influence on the heat production.

"The extra heat which is the product of the specific dynamic action of protein may be used in substitution for the extra heat induced by the effect of environmental cold (Rubner)."

In the case of glucose the demands of working muscles may prevent the development of the carbohydrate plethora. Muscular work, however, has little effect upon the protein metabolism, and the extra heat produced by the ingestion of protein is not available for the performance of muscular work. The energy requirement for work was found by Lusk (175) to be superimposed upon the metabolism induced by the specific dynamic action of protein.

Lusk found that the increase in heat production after ingestion of protein was proportional to the protein metabolism of the time. An increase in the endogenous metabolism of protein, such as occurs in the case of phlorhizin glycosuria is also accompanied by an elevation of metabolism. It will be of interest, therefore, to inquire into the specific dynamic action of protein in those diseases in which occur an increase in the "wear and tear quota" of protein or what is sometimes called a "toxic destruction of protein."

In typhoid fever the minimal metabolism of protein, that is the amount of protein required to repair wear and tear of tissues, is greatly increased. Coleman and DuBois (63) found that the specific dynamic action of protein and carbohydrate is much smaller in the febrile period of typhoid fever than in health and that in some cases it seems to be absent. In convalescence it may be greater than normal.

In *tuberculosis*, on the other hand, McCann and Barr (190) found relatively little evidence of toxic destruction of protein. In this disease the ingestion of protein produces the same degree of increase in metabolism as is observed in health.

Likewise in the *hyperthyroidism* of *Graves' disease* DuBois (75) showed the specific dynamic action of protein and dextrose to be the same as in health, which corresponds with the finding by Rudinger of a normal nitrogen minimum in this disease.

The specific dynamic action of the foodstuffs has a peculiar bearing on the problem of selection of a proper diet in cases of pulmonary tuberculosis. One of the basic principles of all modern therapy of tuberculosis is to secure complete rest for the infected part until healing can occur. This is particularly difficult in the case of pulmonary tuberculosis because complete immobilization of more than one lung is incompatible with life. When the disease is bilateral it is impossible to do more to secure rest for the lungs than to reduce the respiratory movements to a minimum. To accomplish this it is necessary to reduce the metabolism to a minimum for it can be shown that the effective alveolar ventilation varies directly with the total metabolism. On empirical grounds the importance of complete muscular rest during active pulmonary tuberculosis has long been recognized. The rationale of this is undoubtedly explained by the fact that the breathing volume, effective alveolar ventilation, and con-

sequent movements of the diseased lung are least in a patient lying motionless in bed. Other therapeutic procedures directed toward the same end are the control of cough, artificial pneumothorax, reduction of breathing on the involved side by having the patient lie on that side, and the use of weights or sand bags on the chest. Many of the institutions, in which the importance of these measures is fully realized, have, nevertheless, shown great inconsistency in that the patient was roundly gorged with food, no thought being taken of the tremendous stimulation of total metabolism and breathing volume which must be the invariable result. It is true that in later years a reaction has occurred against forced feeding because careful clinical observation revealed many undesirable effects of such a régime (192).

This problem was investigated by McCann (191), who found that when the metabolism was increased by the ingestion of protein or fat a corresponding increase occurred in the effective alveolar ventilation in both normal and tuberculous subjects. When glucose was ingested it was found that the alveolar ventilation was increased to a much greater extent than the total heat production. This result was due to the relatively greater amounts of carbon dioxide which it was necessary to eliminate during carbohydrate oxidation. From experiments of Benedict and Carpenter (36) it was shown that at the peak of the effect of ingesting 100 grams of sucrose the pulmonary ventilation might increase as much as 60 per cent.

McCann (192) found that nitrogen balance could be established in patients with active tuberculosis at a fairly low level. In some cases it was possible to attain nitrogen equilibrium with only 37 to 44 grams of protein in the diet, provided the total caloric values of the diet were from 1.7 to 2.4 times the basal energy requirement.

In order to give such high calorie-low protein diets, large amounts of carbohydrate and fat were required. The fat was found to furnish the maximum number of calories with the least effect upon the breathing volume. The small amounts of protein would produce little effect on the metabolism or respiration, but the large amounts of carbohydrate used would cause a very considerable and undesirable increase in the volume of respiration. This led to the conclusion that such low protein high calorie diets were as undesirable in their way as were the tremendously high protein diets which were once in vogue.

If nitrogen balance is established in tuberculous patients at a level of from 60 to 90 grams of protein the total calories of the diet need not be so high. If this amount of protein is divided into three or more meals the specific dynamic effect of protein at any given time will not be excessive. Furthermore, if advantage is taken of the newer knowledge of antiketogenesis (300, 271) the diet can be made to contain only the minimum amount of carbohydrate which is necessary to insure complete utilization of fat. If one uses Woodyatt's formula (300)

$$F = 2C + P/2$$

a 2500 calorie diet containing 90 grams of protein, 75 grams of carbohydrate and 195 grams of fat, would be more than adequate for a patient of average size with active febrile tuberculosis of the lungs while at rest. Such considerations as the above are obviously of no importance for non-pulmonary forms of tuberculosis.

b. Respiratory exchange and the intermediary metabolism in disease

Since the respiratory quotients which are characteristic of the combustion of carbohydrate (1.0) and of fat (0.71) are so different, the determination of the non-protein respiratory quotient affords a means of calculating the relative amounts of carbohydrate and fat oxidized. In order to calculate the non-protein respiratory quotient it is necessary to know simply the total amounts of carbon dioxide and oxygen exchanged in a given period of time and the amount of protein metabolized during the same period of time. As a measure of the protein metabolism it is customary to use the urine nitrogen excretion during the period of observation. This is at best an approximation because of the fact that there is undoubtedly an interval between the production and excretion of the end products of protein metabolism. This interval probably varies with different individuals and in the same individual it depends somewhat upon the activity of diuresis. The diuresis from water drinking may wash out more nitrogenous end products than are produced in the metabolism of the time. In spite of these objections one may still obtain by the calculation of Zuntz and Shumburg (304) a fairly reliable approximation of the proportions in which the foodstuffs are being oxidized. From the urine nitrogen the amount of protein metabolized is calculated. From this, in turn, a

calculation is made of the amounts of carbon dioxide produced and of oxygen absorbed in the protein metabolism. The carbon dioxide and oxygen of protein metabolism are subtracted from the total amounts of these gases exchanged, the differences represent the CO_2 and O_2 of the *non-protein* metabolism, that is the metabolism of fat and carbohydrate. In this way the *non-protein* respiratory quotient is obtained.

When the non-protein R.Q. is 1.0 all of the heat of the non-protein metabolism is assumed to be derived from carbohydrate. A non-protein quotient of 0.71 would indicate that all of the non-protein calories were produced by fat combustion. Zuntz and Shumburg (304) have calculated the relative percentage of fat and carbohydrate undergoing oxidation simultaneously corresponding to different respiratory quotients between the limits of 0.707 and 1.0.

The heat value of a liter of oxygen consumed in the non-protein oxidation depends upon the relative amounts of carbohydrate and fat undergoing metabolism. When the non-protein R.Q. is 1.0 and carbohydrate alone is being oxidized the heat value of 1 liter of oxygen was calculated by Zuntz and Shumburg to be 5.047 calories. When the non-protein R.Q. was 0.707 and fat alone was burned it was calculated to be 4.686 calories. For intermediate values of the respiratory quotient the heat values lie between the limits given. In this way the heat of the non-protein metabolism may be calculated.

Certain special conditions must be recognized in connection with such calculations. Respiratory quotients, following the ingestion of carbohydrates, are sometimes observed to be greater than one. Provided it can be shown that this is not due to overventilation of the lungs quotients greater than 1.0 may be explained as due to the conversion of carbohydrate into fat. Under special circumstances the respiratory quotient may be less than 0.70. The explanation of such quotients will be discussed presently. Also, it must be recognized that the calculations of Zuntz and Shumburg can not be made if there is reason to suppose that non-protein substances other than fat and carbohydrate are undergoing oxidation. Such conditions arise after the ingestion of alcohols, or during the metabolism of aldehydes, ketones, etc. Finally, it must be emphasized that all such calculations are not valid if the normal control of the breathing is

interfered with in any way, either by the apparatus employed or by the effects of a drug such as epinephrin.

Krogh and Lindhard (151b) have recently advanced an hypothesis, which, if it is substantiated, will introduce new aspects into the interpretation of the respiratory quotient. In work experiments they found that the mechanical efficiency of their subjects was less when the R.Q.'s were low. From their data they calculated that there was a loss of energy amounting to 11 per cent when the energy for work was derived from fat. This waste of energy may be the result of the necessity of converting fat into some other substance which can be used by muscles in performing work. Krogh and Lindhard (151b) regard it is probable that carbohydrate can no more be burned without fat than fat can be burned without carbohydrate. Their hypothesis is that carbohydrate is formed from fat and provisionally stored when the quotient is below 0.8, while a corresponding transformation of carbohydrate to fat takes place when the quotient is above 0.9; that these anabolic processes make the R.Q. lower than the catabolic when this is low and higher when it is high, and that they give rise to an extra expenditure of energy during rest; and, finally, that during work the anabolic processes (combined with storage) are not increased in proportion to the catabolic, whereby the total quotient is lowered when it was high and raised when it was low beforehand.

The above hypothesis best explains the observations of Krogh and Lindhard. It is probably correct as regards the conversion of fat into some other substance before being used as a source of energy for muscular work. The nature of this conversion is still unknown. That fat is convertible into carbohydrate, except for the small amount derivable from its glycerol, is improbable in view of the experiments of Lusk (166), who "found in a phlorhizinized dog which had been rid of glycogen by shivering and exercise that the composition of the urine was unchanged as the result of traveling 1500 meters in a revolving wheel, an effort which would have more than doubled the metabolism of fat during the hour when the exercise was taken." The data of this experiment, given in Lusk's "Science of Nutrition," page 458, are as follows:

	URINARY GLUCOSE	URINARY NITROGEN	D:N
Rest	4.57	1.26	3.63
Work, 1500 meters during first hour.....	4.62	1.26	3.67

In a recent review Shaffer (271) has outlined in an admirable fashion the probable courses of the intermediary reactions occurring during the metabolism of carbohydrates in the body. He points out that, while many interesting facts are known, the fundamental problems remain unsolved. The interesting facts concerning the alterations in the respiratory exchange produced by the ingestion of sugars and of nearly related substances can not yield conclusive information while so much doubt exists as to the nature of the reactions involved.

An enormous amount of work has been done in this field. Lusk (171), performed many experiments in which various sugars were administered to normal and phlorhizinized dogs, the effects upon heat production and respiratory exchange being observed. As regards the intensity of their effects of raising the heat production he found that of fructose most marked, next sucrose, glucose, and galactose, while the effect of lactose was almost negligible. Alcohol was found to increase the metabolism and to diminish the respiratory quotient. Ethyl lactate produced a more marked increase in metabolism than alcohol. Its fate in the body was undetermined since only one-third of the theoretically possible conversion of lactic acid to glucose occurred when the ethyl ester was fed to phlorhizinized dogs.

Lusk's experiments were carried out in hourly periods which do not show the rapidity of the changes in the respiration. Admirable experiments have been performed by Higgins (123), who used an open circuit apparatus to study the exchange at very short intervals. From the diminution of respiratory quotient produced by ethyl alcohol he concluded, that it began to be burned in appreciable quantities in from 5 to 11 minutes after ingestion; that sucrose, lactose and levulose began to be burned quite as soon as alcohol, if not sooner; and that glucose and maltose did not begin to be oxidized until 20 to 30 minutes had elapsed. With sucrose and levulose the respiratory quotient rapidly rose above unity, which phenomenon is usually explained by the conversion of some of the sugar into fat.

In the case of glucose a temporary depression of the R.Q., below the basal level, was noted. This depression may also be observed if one refers to similar experiments reported by Benedict and Carpenter (36). In one of their normal subjects (B. M. K.) ten minutes after the ingestion of glucose a "diabetic quotient" of 0.67 was observed. The significance of such quotients will be discussed later. Some of the experiments of Bornstein and Holm (53) also show a depression of the respiratory quotient for as much as 34 minutes after taking dextrose. Bernstein and Falta (41) have performed experiments which give the most probable explanation as to the cause of this depression of the R.Q. in normal individuals. Normal subjects, to whom was given a diet rich in protein and with little carbohydrate, were made to perform hard work until acetonuria developed. This was done in order to deplete the stores of glycogen. When carbohydrate was given to these subjects it was found that the usual increase in carbon dioxide excretion did not occur. They concluded that the extent of the oxidation of sugar varies with the extent of the glycogen stores.

An observation of a similar nature was made by McCann (189). A normal man, after fasting for eight days, was given a meal of lean beef. During the second and third hours after the meal the respiratory quotients were 0.687 and 0.681, respectively. In the fourth hour the R.Q. was 0.740. The basal values during two hours just before the meal were 0.733 and 0.723. During a control experiment performed one week later, the subject having been on a normal diet in the interval, no significant lowering of the R.Q. was observed. The explanation offered for the phenomenon observed in the first experiment was that as fast as carbohydrate was produced in the metabolism of protein it was deposited as glycogen instead of being immediately oxidized, a depression of R.Q. being thus produced.

Sanger and Hun (264) performed experiments to determine the rapidity of oxidation and mobilization of glucose in hyperthyroidism. Ten patients with hyperthyroidism and ten normal subjects were studied. The excessive mobilization of glucose in hyperthyroidism was shown by the fact that the blood sugar range was twice the normal after ingestion of glucose. The average basal respiratory quotients were lower in the cases of hyperthyroidism and the quotients

rose more rapidly in these cases after taking glucose than in normal persons. These authors calculated that about 18 per cent of the ingested sugar was burned by the normal controls during the two hours after ingestion against 36 per cent by the thyrotoxic cases during the same period. Cramer and McCall (66) came to similar conclusions after studying the respiratory exchange of rats on a standardized diet before and during periods of thyroid feeding.

In phlorhizin glycosuria of dogs Lusk found that administration of glucose and fructose produced little change in the heat production with no evidence from the respiratory quotients to indicate the oxidation of any considerable amounts of these sugars.

In *diabetes mellitus* there have been many interesting studies of the effects of pure foodstuffs on the respiratory exchange. When dextrose is administered to diabetic subjects in many cases a pronounced fall in respiratory quotient is observed, which may be prolonged for several hours. This effect of dextrose was noted in the experiments of Nehring and Schmoll (226), Weintraud and Laves (295) and of McCann and Hannon (194). Experiments are reported by Livierato, Johannsson (136), and Achard and Binet (1) in which the CO_2 excretion was observed to diminish after ingesting glucose, the respiratory quotient not being determined.

The R.Q. is not invariably depressed after giving glucose in diabetes. In some cases no appreciable change in quotient is noted, in others a rise in R.Q. occurs which is less rapid than normal. In two cases observed by McCann and Hannon (194) patients with severe diabetes showed a rise in R.Q. after dextrose which was quite as rapid as that occurring in many normal subjects.

Mohr (213) noted a marked decrease in R.Q. after giving protein to diabetic subjects. This recalls the similar effect of giving protein to a normal man at the end of an eight day fast (McCann 1920). Also, in the subject studied by Wilder, Boothby and Beeler (297) a decrease in R.Q. was noted after meals, particularly if these were rich in protein. In certain mild cases of diabetes Seib (270) noted a rise in R.Q. after protein was given.

The results of administration of levulose to diabetic subjects are often quite different from those produced by dextrose, just as they are with normal subjects. Joslin reports numerous experiments with

levulose. He found that levulose produced an average maximum increase of 0.09 in the R.Q. but that the average R.Q. after levulose was 0.03 above the basal level. Only one experiment with dextrose alone was given, but in this case the maximum rise was 0.03 and the average R.Q. after dextrose was 0.02 above the basal level. Joslin also found that ingestion of orange juice and sucrose were capable of producing considerable elevations of the R.Q. in severe diabetes. A comparison of the ability to oxidize dextrose and levulose was made by Wilder, Boothby and their coworkers (298). These authors also found that levulose produced an elevation of R.Q. much more readily than glucose in diabetic subjects, both before and after treatment with insulin, confirming the view long held by Minkowski regarding the superior utilization of levulose.

On account of the importance of phosphoric acid in the intermediary metabolism of carbohydrate it is of interest to compare the effects of dextrose and of the hexose phosphoric ester on the respiratory exchange in diabetes. Such a comparison was made by McCann and Hannon (194) who found that the respiratory quotients rose more readily after the ingestion of calcium hexose phosphate than they did in the same diabetic subjects after ingestion of dextrose. The hexose phosphoric ester on hydrolysis yields a levo-rotary reducing body which is probably levulose. The formation of levulose from the hexose phosphate may account for the more rapid elevation of the R.Q. in the diabetic subjects whose power of oxidizing glucose was impaired.

Glycerol was found by McCann and Hannon (194) to produce a change in the R.Q. of diabetic subjects quite similar to that produced by dextrose. Glycerol however produced a fall in R.Q. in normal subjects. When glycerol and glycerophosphates were compared as to their effects on the respiratory metabolism in diabetes no evidence of a superior utilization of the phosphoric ester was obtained.

Concerning the *significance of the respiratory quotient* in diabetes there is much uncertainty. The vast majority of diabetic subjects are found to have quotients much below the average of normal subjects in the post absorptive condition. In a large series of observations by Joslin the average diabetic R.Q. before June, 1914 was 0.73, and after that date it was 0.77. In general it is safe to assume

that the quotients are low because of the very small portion of the calories derived from carbohydrate. A tendency for the R.Q. to rise is generally welcomed as evidence of increasing ability to utilize carbohydrate. Allen and DuBois (2), and Joslin (138) point out extremely high respiratory quotients are sometimes encountered in severe diabetics (R.Q. = 0.94) under circumstances which make it highly improbable that carbohydrate is being oxidized. Joslin is inclined to interpret these high quotients as indicating a conversion of carbohydrate into fat. Such cases observed by him were usually in extremis and greatly underweight and proved to have very low basal metabolism. The lack of available fat may have encouraged the formation of fat from the superfluous sugar. Concerning other factors which tend to elevate the R.Q. in diabetes one must consider the oxidation of acetone bodies and the excretion of a high proportion of urinary nitrogen as NH_3 instead of urea.

Abnormally low respiratory quotients (below 0.707) are encountered with great frequency. Lusk (170) has written the most adequate discussion of the probable significance of these low quotients. He calculates that if none of the carbohydrate, which is formed from protein is oxidized, the R.Q. for protein would become 0.63 instead of 0.81. He quotes Magnus Levy's calculation that the formation of the maximum amount of β -hydroxybutyric acid from fat would change the quotient for fat from 0.707 to 0.669, but Lusk adds that the β -hydroxybutyric acid acting on bicarbonates in the body would displace CO_2 with the result that the quotient would be elevated rather than depressed. However such extremely low quotients are practically never encountered except when marked acidosis exists. It is quite obvious that the interpretation of the respiratory quotient in diabetes is largely speculative at the present time, and that existing views may be much modified by the conceptions of Krogh and Lindhard (151b).

Joslin (138) states that "the respiratory quotient in diabetes appears to bear a definite relation to the metabolism in that it is low when the metabolism is high and higher when the metabolism is low." This statement is in accord with the findings of Krogh and Lindhard (151b), who found that the standard metabolism was lowest at intermediate quotients and higher with very low quotients and with very

high $R.Q.$'s. The variations of *heat production at different quotients* noted by Joslin (138) are greater than those noted by Krogh and Lindhard, whose subjects had a minimal protein metabolism. For instance Joslin states that "the average respiratory quotient before June, 1914 was 0.73 and after June, 1914, 0.77. The average metabolism was 13 per cent *above* standard in the earlier period and 10 per cent *below* standard in the later period." Prior to June, 1914, Joslin's patients had been treated by older methods which permitted the use of large quantities of protein and fat. After that date the method of fasting and undernutrition was used with superior results. Wilder, Boothby and Beeler (297) have also observed that clinical improvement is accompanied by a decrease in the metabolic rate. In the author's own series of cases this observation has been confirmed, and it is his opinion that the decrease in metabolism occurs when the endogenous protein metabolism decreases as the result of low protein intake and increased utilization of fats and carbohydrates. It is notable that the nitrogen excretion per kilogram of body weight of Joslin's patients before June, 1914, was almost one half greater than the nitrogen excretion after that date. The coincidence of marked decrease in basal metabolism with clinical improvement and decreasing protein metabolism is beautifully illustrated in the case of Gerald S. studied by Allen and DuBois (2).

Calorimetric observations have been used in a variety of ways in studying the effects of different types of treatment. Allen and DuBois (2) studied the "oatmeal treatment," which was once in vogue. In their observations no special influence of oatmeal in diabetes or special readiness of oxidation of this form of carbohydrate was demonstrable. The respiratory exchange failed to account for all the carbohydrate that disappeared.

McCann, Hannon and Dodd (196) showed that the improvement of patients treated with insulin was very well brought out by comparison of the curves of the $R.Q.$'s after ingestion of glucose before and during the course of treatment. Patients in whom glucose at first caused depression of the $R.Q.$ subsequent to a period of treatment with insulin showed curves in which the quotient rose promptly.

In the foregoing review of the contributions of the study of respiratory metabolism to the problem of diabetes mellitus the author

acknowledges very numerous omissions. It is impossible at the present time to arrange in a coherent and orderly manner all of the immense number of experiments in this field. It is believed that the main trends of investigation have been presented.

PART III. THE MECHANICAL EFFICIENCY

The cost of muscular effort in pathological states has received scant attention from clinical investigators. It is most unfortunate that so little has been done in what is, after all, a very important field.

Using the nitrous oxide method of Krogh and Lindhard it was shown by Boothby (46) that the rate of circulation of the blood and the pulmonary ventilation varied proportionally with the rate of oxygen consumption in normal subjects. Means and Newburgh (199) found a progressive increase in blood flow in response to increasing amounts of work. R. G. Pearce (237) has also given a most interesting discussion and data bearing on the same points. The technical details of these investigations need not be discussed in this place. The important fact is that the functional demands made on the respiratory and circulatory mechanism depend upon the rate of metabolism. It is highly essential in the rational management of patients with respiratory and circulatory diseases that the physician should have some idea of the effect of muscular effort upon the rate of metabolism.

In exophthalmic goitre the occurrence of tachycardia is one of the cardinal phenomena of the disease. Sturgis and Tompkins (281) have shown very strikingly the parallelism of the changes in the basal pulse rate and the basal metabolism in hyperthyroidism. It would seem that the acceleration of the pulse in Graves' disease gives a fairly accurate measure of the increased rate of circulation of the blood required by the increased metabolism at rest in the majority of patients. Careful observation of severe cases of Graves' disease reveals the fact that breathlessness on slight exertion is a very common symptom. In the later stages of the disease evidences of myocardial damage, arrhythmias, especially auricular fibrillation, are frequently observed. It is quite clear that the functional demands made upon the heart muscle in Graves' disease are very great and that in many cases severe myocardial injury is present. Striking examples of

myocardial lesions in hyperthyroidism have been given by Goodpasture (100), who found that products of the thyroid alone could not be entirely responsible for the cardiac lesions. Whatever may be the cause of the increased susceptibility to injury of the heart in hyperthyroidism the increased functional demands upon this organ must be an important contributory factor.

An entirely new phase of this subject has recently been investigated by Plummer and Boothby (246). These authors compared the mechanical efficiency of six patients with exophthalmic goitre with that of three normal men and four debilitated patients. The average net expenditure of energy of the normal and debilitated individuals was 1.18 calories per kilogram meter of work performed in walking horizontally on a treadmill at the rate of 800 meters per hour. The average net cost of the same work by the exophthalmic patients was almost twice as great, being 2.22 calories per kilogram meter. Furthermore it was brought out that the mechanical efficiency may increase if the basal metabolism is decreased following operative treatment. Thus one of these patients whose metabolism was 51 per cent above the average normal required a net expenditure of 2.07 to 2.16 calories per kilogram meter. Six days after operation the basal metabolism was only 25 per cent above average normal and the net cost of the same work was only 1.73 to 1.75 calories. These findings are most significant when one considers that every effort on the part of a patient with hyperthyroidism may require twice as great an increase in blood flow and in alveolar ventilation as that which would occur in a normal individual in performing the same external work.

Similar investigations had previously been made by Peabody and Sturgis (236) on patients with organic heart lesions. It was found that the slight amount of exercise involved in climbing up sixty steps produced the same relative changes in oxygen consumption, pulmonary ventilation and heart rate in a group of patients with heart disease and in a similar group of normal individuals. The greater subjective dyspnoea of these patients does not depend upon a greater metabolic cost of work, but rather upon secondary alterations in the respiratory mechanism.

Curiosity is immediately aroused as to the possible bearing of similar studies in the group of individuals, who suffer from breath-

lessness, palpitation, tachycardia and excessive fatiguability without demonstrable lesions in the circulatory or respiratory organs. In these cases various diagnoses have been applied such as Effort Syndrome, Disordered Action of the Heart, and Soldier's Heart. Are these symptoms due to low mechanical efficiency so that the performance of work is accomplished at the cost of an abnormally great expenditure of energy causing an excessive response on the heart of heart and lungs? At present no data are available, though the problem is undergoing investigation in the author's laboratory.

It will be seen from the foregoing account that very little is known of the mechanical efficiency of the body in disease. For the benefit of those who desire to investigate this interesting subject there exists an abundance of physiological studies which may serve as a guide to the technique of investigation and as controls to the results obtained in pathological states.

The early physiological literature concerning muscular efficiency has been well reviewed by Benedict and Cathcart in their monograph on muscular work (31). A more recent review of the literature may be found in Lusk's classical work on the Science of Nutrition. This latter author has dealt with the subject in the widest sense, being particularly concerned with qualitative as well as quantitative changes in the metabolism during muscular work.

The mechanical efficiency of normal individuals varies with the nature of the work. Differences exist between efficiency in bicycle riding and in walking, and between walking on a level and walking up an incline. Even in horizontal walking the expenditure of energy in walking on a laboratory floor may be much less than in walking on a level grass plot at the same rate. (Douglas, Haldane, Henderson and Schneider (69).)

The effect of training on the efficiency of subjects is most important. Unfortunately for the clinical investigator a great deal of the physiological work has been carried out with trained subjects. In the study of abnormal subjects controls should be selected who have not undergone special physical training. Special training for a particular kind of work may also increase efficiency in performing that work. Of two individuals whose efficiency in walking is equal, one might prove to be much more efficient in bicycle riding (through practice)

than the other. For clinical trials horizontal walking at low rates of speed would seem to be the best form of work because of the fact that practically everyone is accustomed to walking.

The efficiency of muscular work depends to a great extent upon the rate at which the work is performed. Lupton (165) caused his experimental subjects to ascend stairs at different rates of speed. He found that the mechanical efficiency of movement varies with speed in a manner corresponding to that predicted by A. V. Hill (128).

The effect of diet upon the mechanical efficiency is of considerable importance. In 1901 Frentzel and Reach (91) in the laboratory of Zuntz showed that the extra metabolism per unit of work was greater when fat was the principal source of energy than when carbohydrate was the chief source. Anderson and Lusk (4) found that a dog could perform work more economically after glucose ingestion than it could during starvation or after the ingestion of meat or alanine. The experiments of Lusk show that very little if any of the energy derived from protein is available for muscular work.

Krogh and Lindhard (151b) have performed a most careful series of experiments during muscular work from which they have shown that there is a considerable waste (about 11 per cent) when fat is the source of energy. In these experiments the proportions of foodstuffs utilized was calculated from the respiratory quotient. The general character of the metabolism during the experimental periods was regulated by means of the diet. A most important feature of the diet regulation was the establishing of a minimal nitrogenous metabolism so that the specific dynamic effect of protein would not be a disturbing factor.

It is clear that work can be more economically performed when carbohydrate is the chief source of energy. The differences in mechanical efficiency on different diets is less than any differences which will be found to possess clinical significance. It will be well for experimenters to establish the same conditions as to type of diet preceding efficiency tests for both normal and pathological subjects, in order to eliminate minor differences due to diet.

The effect of altitude and of oxygen tension upon the muscular efficiency may enter into the interpretation of results obtained in some pathological states. The members of the Pikes Peak Expedition

(69) found no great differences in the metabolic cost of walking on Pikes Peak after acclimatization and at sea level. Nor did Hasselbalch and Lindhard (120) detect a difference in the cost of work in a cabinet at low oxygen pressure. E. C. Schneider (268) has reported recently that physical exercise caused a higher rate of metabolism during the first days at 14,110 feet than was normal at lower altitudes.

PART IV. TOTAL ENERGY REQUIREMENTS IN DISEASE

From the foregoing discussion it must be evident to the reader that calorimetric observations of normal men provide insufficient data for the calculation of the energy requirements in disease. In any given pathological state the energy requirement may be said to be the amount of energy necessary to maintain the weight and to prevent waste of body protein, and in the case of children, to permit growth as well. It concerns not only the total energy value of the food, but also the proportions in which energy is derived from the foodstuffs, protein, fat, and carbohydrate.

Fevers

In typhoid fever Shaffer and Coleman (272) found that it was necessary to employ diets furnishing a large number of calories in order to establish nitrogen balance and weight equilibrium. Coleman and DuBois (63) found that the basal metabolism in typhoid fever was increased from 36 to 40 per cent above the average normal. It was also observed that it was necessary to give a diet which exceeded the theoretical energy requirement by from 50 to 110 per cent in order to bring patients into weight and nitrogen equilibrium (64). The administration of this excess of food did not appear to increase the basal metabolism, so that the fate of the excess food has never been determined. There was some evidence that typhoid patients could store fat in the body even while losing body weight and body protein. The satisfactory elucidation of this point would require continuous observation of a patient receiving a high calorie diet throughout a period of twenty-four hours.

A somewhat similar state of affairs concerns the total energy value of the diet required in tuberculosis. McCann and Barr (190) found

no increase in the basal metabolism in tuberculosis during afebrile periods. With the elevation of temperature an increment in metabolism was observed, but the size of the increment in terms of the daily heat production depended upon the duration of the fever. Many tuberculous patients are afebrile for a considerable portion of each twenty-four hours. In subsequent studies McCann found that it was necessary to furnish the patient a diet containing from 1.7 to 2.4 times the theoretical basal energy requirement, in order to establish nitrogen equilibrium at a low level (37 to 45 grams of protein daily). When nitrogen equilibrium was established at a higher level a smaller number of total calories in the diet was required. The decision as to which type of dietary regimen will yield the best results in the treatment of tuberculous patients must be made only after careful clinical and metabolic study of a large number of patients for a considerable period of time. There is evidence of unfavorable effects from high-calorie diets in pulmonary tuberculosis during periods when activity of the disease makes it necessary to restrict all muscular effort. It is highly desirable that further work be done to determine the metabolic cost of work in pulmonary tuberculosis especially in reference to the rate of blood flow through the lungs and the rate of ventilation of the lungs.

Food requirements of children

The dietary needs of the normal child are matters of prime concern to the physician. During recent years the nutrition of infants has received a great amount of attention resulting in a much more satisfactory knowledge of the energy requirements, mineral necessities, and "vitamines" essential for normal activity and development. Far less study has been given to the equally important needs of children from two to six years, and of those of school age.

The problem has been attacked from the standpoint of calorimetry by workers like Benedict and Talbot (38) and others who have contributed so much to our knowledge of the basal metabolism of children (220, 132, 219, 287). It is not possible to solve the problem entirely by such means. The food requirement of a child is determined by four factors: the basal metabolism, the amount of food lost in the feces, the quota for growth, and the degree of muscular activity.

Attempts have been made to study children under various conditions of activity in respiration chambers (280), etc., but this method does not furnish the essential knowledge of the growth quota, nor does it really enable one to study all of various forms of activity of a vigorous healthy child.

A most valuable method was employed by Gephart (97) in a study of the actual food consumption of boys from thirteen to sixteen years of age in St. Paul's School, Concord, N. H. These boys were found to consume daily about 5000 calories, or about three times the basal requirement. Estimates were made from a knowledge of food stores consumed, food waste in garbage, with a careful tally of the number of individuals served. Average figures only were obtained. The basal metabolism was estimated from measurements of the height and weight of the boys, using average normal figures obtained from DuBois' laboratory. This method was subsequently employed by Murlin in his valuable studies of the army ration.

Holt and Fales (131) have made most complete and careful studies of the spontaneous intake of one hundred healthy children of various ages, determining in this way the energy value of diets which were adequate for maintenance of normal growth and activity. The following table gives a summary of the average findings of the energy needs of children of various ages. Individual variations were rather large due to variations in muscular activity.

Energy requirements of children (Holt and Fales (131))

	<i>calories per kgm.</i>
1 year (under 9.5 kgm.).....	100
Boys, 6 to 15 years (20 to 50 kgm.).....	80
Boys, 15 years to adult.....	48
Girls, 6 to 10 years.....	76
Girls, 10 years to full growth.....	80
Girls, growth complete.....	44

Holt and Fales (131) found a fairly uniform loss in the feces of ten per cent of the total intake. It was also determined that the percentage of calories derived from the various foodstuffs was quite uniformly distributed as follows:

	AVERAGE	RANGE	GROUPING
	<i>per cent</i>	<i>per cent</i>	
Protein.....	15	11-23	$\frac{1}{2}$ between 14 to 16 per cent
Fat.....	34	21-51	$\frac{3}{4}$ between 30 to 40 per cent
Carbohydrate.....	51	38-65	$\frac{3}{4}$ between 44 to 55 per cent

An important study of the spontaneous intake of infants from the fifth to twelfth day has also been made by Faber (87), in which it was determined that the actual intake was in excess of the estimates based on calorimetric studies.

The foregoing investigations are of enormous value from the standpoint of preventive medicine. They will also form the starting point of much further investigation of the energy requirements in diseases of children, concerning which little is known at present.

RÉSUMÉ OF THE CONTRIBUTIONS OF CALORIMETRY TO MEDICINE

As a result of calorimetric studies the basal metabolism of normal adults may be predicted with a fair degree of accuracy. That of normal children is likewise predictable, though less accurately than with adults. The basal metabolism of normal adults may vary from time to time over a range of ± 15 per cent of the average. Environmental influences and nutritive conditions are factors in these normal day to day variations.

The development of normal standards for basal heat production made possible the determination of variations from normal in pathological states. The diseases, in which gross variations from normal were discovered, are those of the thyroid gland, the fevers, and certain diseases of the hematopoietic system. In those conditions which produce marked abnormalities of the basal metabolism its determination for diagnostic purposes is usually superfluous, though it remains of value as a measure of the severity of hyper or hypothyroidism, and is thus useful as an index of the effectiveness of treatment of these two conditions.

Minor variations from the average normal basal metabolism occur in such a wide variety of pathological states that a searching clinical analysis is necessary for the evaluation of the metabolism test. The

possibilities of technical errors in the determinations are greatly multiplied by the widespread use by inexperienced observers of simple forms of apparatus lacking adequate checks and controls. Not all of the errors lie within the apparatus. Careful control of environmental and nutritional conditions and avoidance of emotional reactions are called for. Taking all things into consideration the diagnostic value of basal metabolism tests may be said to be very slight.

Calorimetry has made notable contributions to the knowledge of the heat regulating mechanisms in fever, to the action of antipyretic drugs, and to the knowledge of dietary requirements in fevers. It has added much concerning the actions of drugs, notably of extracts of thyroid, adrenals, and pancreas.

The study of the respiratory exchange has greatly elucidated the intermediary metabolism in health and in disease, particularly in diabetes.

The problem of determining the food requirements in health and in disease has not been solved entirely by calorimetry. One may prescribe a diet on the basis of calorimetric data, but "the proof of the pudding is in the eating." In some instances the energy requirement has been determined by measurement of the food intake which was required to maintain normal weight, to spare body protein, and, in the case of children, to promote normal growth. The use of this method has in several instances proved the energy requirement to be greater than the prediction based on calorimetric measurements alone, for instance, in the case of typhoid fever and of normal children. The method is, however, a special application of calorimetry, since a knowledge of the heat values of foods is essential to the computation.

The interrelationships of the energy transformations with the functional demands on the heart and lungs are important considerations in regulation of the diet and exercise, for instance, in such a disease as pulmonary tuberculosis. When further studies of the mechanical efficiency have been made a knowledge of these relationships will find important therapeutic applications in many diseases.

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A REVIEW OF THE INVESTIGATIONS CONCERNING THE ETIOLOGY OF MEASLES

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INTRODUCTION

The etiology of measles and, indeed of all the exanthemata of man, constitutes one of the perplexing problems in medicine. At one time, the somewhat discouraging impression prevailed that these infections are probably caused by a type of parasite wholly unlike any of the pathogenic micro-organisms with which we are familiar; on the other hand, an opinion, decidedly too hopeful, has sometimes been expressed that the discovery of the etiologic agent of one of the acute exanthemata would lead to the clearing up of the entire group. A little reflection shows that this latter view is rather sweeping. The underlying pathological processes of the various exanthematous lesions are often dissimilar and micro-organisms as diverse as (1) *Treponema pallidum*, (2) *Rickettsia prowazeki*, and (3) *B. typhosus*, are each capable of producing a well marked skin rash.

In the investigation of infectious diseases of unknown origin, the clinical features frequently suggest the general group of parasites in which the etiologic agent would most likely be found. In the study of measles, it is important to note its mode of transmission, the portal of entry of the virus, the self-limited course of the infection and the resulting immunity. Occasionally, investigators have described protozoan-like bodies as the causative agent of the disease, apparently disregarding the consideration that typical protozoa are not transmitted by droplet infection and usually produce a more or less chronic type of disease which is not followed by a sterilizing immunity. It would be extremely surprising if the etiologic agent of measles should prove to be a protozoan parasite.

In reviewing the experimental work concerning measles, three general subjects will be considered, namely (1) the histologic pathology of the specific lesions, (2) the artificial inoculation of the disease in man and lower animals, and (3) cultural studies on artificial media. These procedures represent the underlying principles upon which the study of an unknown virus is usually undertaken. Various investigators have studied these features rather extensively in measles with results that are sometimes difficult of interpretation, but which possess considerable interest. The divergent observations described in the literature will often necessitate the introduction of considerable detail in this review in order to bring these conflicting data into harmony as far as possible.

HISTOLOGICAL INVESTIGATIONS

The chief interest in the microscopic examination of the lesions of measles in the skin and mucous membrane lies in the search for the specific etiologic agent. Since the portal of entry and the primary lesions of measles occur in the mucous membrane of the respiratory tract, it seems reasonable to assume that the Koplik spots are produced by the specific organism of the disease *per se*. The clinical features indicate that the exanthem is caused by the presence of a specific microorganism and not solely by a toxin. The generalized rashes caused by agents in solution, such as drugs and sera, make their appearance irregularly over the surface of the body. Areas that are widely separated may become involved simultaneously. In measles there is a definite and rather gradual progress of the rash from the uppermost parts of the body downward over the trunk, arms, and lower extremities.

Careful microscopic examinations for parasitic organisms has revealed very little, either in the Koplik spots of the mucous membrane or in the skin lesions. In the endothelial cells of the capillaries running through the skin lesions and also in Koplik spots, Mallory and Medlar (1) found coccoid bodies of variable size occurring in the cytoplasm, either singly or in pairs. These bodies stained positively by Gram's method. Their exact nature could not be determined. The authors considered it unlikely that these bodies could

be the remains of phagocytosed and digested leucocytes. Neither did it seem probable that they were retrograde changes since these cells tended to proliferate rather than degenerate. These bodies in some respects were similar to enlarged centrosomes but the authors considered that they were probably cocci in various stages of dissolution. They were found in the digestive vacuoles in the cytoplasm and they did not occur in the blood vessels in unaffected tissues of the body. Nothing resembling these bodies could be found in control examinations of a variety of other skin lesions.

A thorough examination of fresh and stained blood smears for parasites was made by Mallory and Medlar in 60 instances but with negative results. In 15 cases, examinations were made every four hours beginning twenty-four to forty-eight hours before the eruption started and continuing twenty-four to thirty-six hours after its appearance. This failure to recognize microorganisms does not by any means preclude their presence in the skin lesions. One need only recall the difficulty not infrequently encountered in demonstrating microscopically in tissues such organisms as *B. tuberculosis* or *T. pallidum*.

The histological pathology of the lesions of measles becomes of direct interest in view of its possible bearing upon the diagnosis of experimental measles in lower animals.

The older writers considered that the roseolae of measles, developed principally around the sebaceous glands, the sweat glands, and the hair follicles. It is definitely shown by Mallory and Medlar that the lesions commence around the capillaries and venules in the skin, starting at a given point and spreading along these vessels. Incidentally the glands and hair follicles become secondarily involved. Their findings may be stated very briefly as follows:

The essential cellular reaction of the skin lesions in measles may be summed up as consisting of a proliferation of vascular endothelial cells, of an emigration of endothelial leucocytes and of a rapid proliferation of them in the tissues surrounding the vessels. The endothelial cells become swollen and finely granular and show occasional mitoses. The endothelial leucocytes are young and active, showing numerous mitoses. They are often larger than normal and they phagocyte and digest the occasional polymorphonuclear leucocytes which wander into the infiltrated area.

Epithelial structures, adjacent to the lesions become infiltrated with endothelial leucocytes. In all early lesions, collections of exudate were noted in the skin beneath the cornified layer, containing serum, fibrin and endothelial leucocytes. No evidence was found of any necrotic lesions such as the pyogenic cocci produce. Very little diapedesis of red cells was observed.

The authors considered that these findings signify that the micro-organisms of measles is phagocyted by the endothelial cells of the capillaries and venules of the skin and mucous membrane, producing an inflammatory reaction in the immediate neighborhood.

It is significant that, in an excised typhoid rose spot, the authors report "a cellular reaction like that of measles, but rather more abundant."

Ewing (2) in 1909, in describing the lesions of measles, noted the mitotic figures in the endothelium and the extensive infiltration of the lesions of the mucous membrane and of the skin with large mononuclear leucocytes. No single type of cellular change was found constantly. Indeed, some peculiar changes observed in a case of hemorrhagic measles were duplicated almost exactly in a case of *pityriasis rosea*. Ewing considered that, from the histological picture alone exclusive of clinical considerations, one might think that the diagnosis of measles included more than one disease. He surmised that measles is caused by an actively multiplying microorganism, of the class of bacteria, producing an active toxin having a special affinity for superficial epithelial cells.

Field (3) in 1905, studied some of the supposedly protozoan micro-organisms which had been described in measles. He concluded that these bodies were not protozoa, but that they were probably degenerating and cytolyzed epithelial cells and leucocytes, which within certain limits are specific for measles and scarlet fever.

The significant and at the same time unfortunate feature of the preceding observations lies in the fact that no pathognomonic cellular reaction in measles has as yet been clearly established. Therefore in studying sections of doubtful rashes such as may occur in experimental animals, one may determine whether the lesions are consistent with measles but an absolute diagnosis can not be established solely from the histological picture.

ARTIFICIAL TRANSMISSION

Inoculation of man. The early experiments on the artificial infection of man with measles were unfortunately conducted without adequate precautions for avoiding the possibility of accidental infection. Hektoen (4) has compiled a thorough review of this early work.

In the experimental study of measles, it becomes of the utmost value to the investigator to know whether the virus is present in the blood stream. From the pathology of the disease, it is self-evident that the causative organism is not invasive to the extent of setting up lesions of the viscera. The decidedly mechanical nature of the progress of the exanthem over the body at once raises doubt about the presence of the virus in the circulating blood.

In 1905, Hektoen (5) conducted the first modern work under carefully controlled conditions upon the experimental inoculation of measles in man. Two volunteers, injected with blood of measles patients, developed symptoms of measles after an incubation period of ten to twelve days. The details of this experiment are very important. The first subject was inoculated with a specimen of blood taken from a patient about six hours after the first appearance of the rash. In order to detect any secondary invaders, the blood (3 cc.) was incubated in ascitic broth (50 cc.). After twenty-four hours there was no gross or microscopic evidence of any micro-organisms and the first volunteer was injected subcutaneously with 4 cc. of this blood, diluted in broth. The quantity of patient's serum injected was approximately 0.1 cc. No local symptoms appeared at the site of injection. Twelve days later, the temperature commenced to rise, reaching 104°F. on the fourteenth day. During the morning of the fourteenth day, a red papular eruption appeared on the forehead and spread over the greater part of the body in about five hours. During convalescence a branny desquamation appeared. There were no catarrhal symptoms and no definite evidence of malaise.

The blood for the inoculation of the second subject was taken from a patient thirty hours after the appearance of the eruption. After incubation in ascitic broth for twenty-four hours, several cubic centimeters were injected subcutaneously as in the preceding case without producing any local reaction. The temperature began to

rise ten days later, reaching 103°F. on the twelfth day. On the following day, a rash appeared on the face spreading to the chest, back and abdomen. No mention is made of any subsequent desquamation. Mild respiratory symptoms, a little conjunctivitis, and slight malaise were present in this patient.

The incubation period observed in these two cases of experimental measles corresponds to that of the spontaneous disease. The exanthem is described as typical of measles. Moreover, the rash appeared first on the face as in spontaneous cases, a point which is of interest in view of the altered portal of entry of the virus.

There are several respects in which the symptoms differed from the ordinary course of the disease. The description in the first case of the rapid progress of the rash over the body within a few hours is quite unlike the usual slow progression as seen under natural conditions. Apparently no desquamation occurred in the second case. The absence of conjunctivitis and of respiratory signs does not, as suggested by Ustvedt (6) militate against the diagnosis of an inoculated form of measles. Neither case showed a pre-eruptive rise in temperature, a feature, however, which is not constant in spontaneous infections. It is unfortunate that no information is supplied in these cases concerning the leucocyte count, the occurrence of Koplik spots and the behavior of the lymphatic glands.

Hiraishi and Okamoto (7) in attempting active immunization against measles, inoculated 44 children with blood from early cases. They conclude that the minimum infective dose lies between 0.001 and 0.002 cc. and that 0.0001 cc. of blood is harmless. The work was carried out during an epidemic. It is by no means clear that adequate precautions were taken to protect these children from accidental infection. No description is given of the experimental disease.

During the winter of 1918 to 1919, the writer (8) inoculated a series of volunteers with blood from early cases of measles in an effort to confirm Hektoen's results. In working with such a common infectious disease, considerable difficulty was experienced in obtaining susceptible adults. Eight volunteers were eventually accepted who, as far as could be determined from correspondence with their families, had never been exposed to measles. These men

were injected in various ways with blood but no symptoms developed in any instance.

The description of these injections may be summarized as follows: For the first inoculation, blood was taken from a patient twelve hours after the eruption appeared. The serum was separated by centrifugalization and diluted with nine parts of isotonic salt solution. One individual was given 5 cc. of the diluted serum subcutaneously.

For the next series of inoculations, a specimen of blood was taken from a case of measles twelve hours after the rash appeared. A portion of this specimen (4 cc.) was incubated in ascitic broth (50 cc.) according to Hektoen's technique and another part was defibrinated. The latter was injected at once subcutaneously in 2 cc. quantities into each of 2 men. The portion in ascitic broth was incubated for one day and 10 cc. quantities were injected subcutaneously into 2 individuals.

Since no symptoms followed the preceding inoculations, some more intensive injections were carried out. Blood was taken in citrate from 2 cases of measles in the pre-eruptive stage, six hours before the rash appeared in 1 patient and thirty hours before its appearance in the other. These citrated specimens were mixed and the equivalent of 3 cc. of blood was injected into each of 2 individuals, part of the injection being given subcutaneously and part intramuscularly. Twenty-four hours later each of the 2 volunteers received a second injection from these 2 patients in the same manner. One of the measles cases was now in the eruptive stage and in the other the rash appeared six hours later. One of these two volunteers gave an unusually clear history of susceptibility to measles. He was the sixth of 8 children and had always lived on an isolated farm in West Virginia. According to the statement of the mother and eldest sister, measles had never occurred in the household. But several members of the family had left home and eventually had contracted measles. Of the older brothers and sisters, 4 out of 5 developed the disease away from home. Of the two younger children, one, a brother, enlisted in the army and developed measles at Camp Shelby, Miss.

Neither of these 2 individuals receiving intensive injections from patients in the pre-eruptive and eruptive stages developed any

symptoms. After an interval of three weeks, they were exposed to an early case of measles and also inoculated on the mucous membrane with secretions from this case in the pre-eruptive stage, four days before the rash developed. The volunteers remained free from symptoms. This result, therefore, suggests that they were immune to measles at the time this final test was made. It is not possible to determine definitely whether their immunity may have been due to some previous unremembered or undiagnosed attack of the disease, or whether it resulted from the injections of measles blood which they received. Certainly the evidence of their susceptibility at the beginning of these injections is more concrete than the generalization that few adults have escaped an attack of the disease in childhood.

Finally, an injection was made in 1 volunteer with whole blood taken from a patient six to twelve hours after the rash appeared. Immediately after withdrawal, without the use of citrate, 0.5 cc. was given subcutaneously and 1.5 cc. intravenously. He remained free from symptoms.

These 8 successive failures indicate that measles cannot be transmitted by the injection of patient's blood as readily as would be expected from the results of the 2 cases reported by Hektoen. Moreover, a thorough analysis fails to suggest any simple or definite explanation of these divergent results. Except in 2 cases, the technique which I followed differed from that of Hektoen, the blood from the measles patient being injected directly without preliminary incubation. At the time these experiments were conducted, it was thought that the direct injection of a moderate amount of blood would be more likely to infect than the use of a minimal quantity after twenty-four hours incubation. Hektoen used approximately 0.1 cc. of patient's serum. However, it is theoretically possible that multiplication of the virus of measles may have occurred during the incubation. If such development did take place, then the preliminary incubation would surely enhance the possibility of reproducing the disease.

One must consider the possibility of producing a fever and rash by toxic constituents contained in the media which was injected. The writer has carried out injections of ascitic broth incubated with normal blood in a series of 20 individuals. Only minor reactions

developed and they could not in any way be confused with the symptoms of measles.

The evidence presented by Hektoen indicates that the fever and the accompanying rash, developing after a period of two weeks constituted true infections with the virus of the disease. Careful precautions were taken to guard against accidental infection during the period of experimentation. Although the resulting symptoms did not conform fully with the naturally acquired disease, it is not to be expected that the injection of a virus under highly artificial conditions would reproduce, in detail, the usual symptoms of the natural infection. The absence of a pre-eruptive rise in temperature, the rapid spread of the rash over the body, the lack in 1 case of inflammation of the mucous membranes, and the very moderate degree of malaise might readily be accounted for by the artificial mode of inoculation.

It is perhaps natural to feel that the blood of a measles patient taken early in the disease would either consistently fail to infect or else regularly reproduce the disease upon injection in a susceptible individual. Such an assumption, however, is not justifiable as a general conclusion. Indeed, the blood of an active case of pneumonia or of typhoid fever, during the stage of bacteremia, might give very inconstant results upon injection into susceptible individuals. The failure in my own work to produce measles in volunteers by the injection of the blood of patients cannot, in my opinion, be explained merely on the supposition that the apparently susceptible volunteers were in reality immune on account of some previous attack of this disease. It is entirely possible that the blood of measles patients, even though the virus be present, would not consistently infect susceptible men. Hektoen's successful results are very important in demonstrating that the virus is present in the blood and that infections can be produced in man by the subcutaneous route even though the normal portal of entry is by way of the mucous membranes. It would be extremely important to know whether the likelihood of successful infection is increased by the preliminary incubation of the patient's blood in ascitic broth as practised by Hektoen. Unfortunately, the results of my experiments throw no light on this question.

Inoculation of monkeys. Experiments upon the transmission of measles to lower animals have been carried out extensively with monkeys, principally those of the genus *macacus*, the injections having been made with blood and mucous secretions of measles patients. Following Hektoen's work with volunteers, Anderson and Goldberger (9) reported the successful inoculation of monkeys in a manner analogous to the production of typhus fever in lower animals. Subinoculation through a series of monkeys produced mild symptoms which these authors interpreted as a reaction to the virus of measles. Confirmation of this work has been reported by several observers though the results of the individual investigators vary rather markedly. One would hardly expect that the typical clinical features of measles could be reproduced in monkeys with sufficient clearness to permit a diagnosis from the symptoms alone. It would be sufficient to produce a perfectly definite reaction which, by the exclusion of other factors, may be proved to be caused by the virus.

There are in all six signs or symptoms which have been reported in monkeys; namely, (1) fever, (2) rash, (3) Koplik spots and other forms of enanthem, (4) leucopenia, (5) conjunctivitis and rhinitis, and (6) evidence of malaise.

Anderson and Goldberger employed three species of monkeys, namely, *M. rhesus*, *M. cynomolgus*, and *M. sinicus*, using in all, more than 100 animals. Apparently these three species were equally satisfactory, though the symptoms were very mild and many individual animals failed to react. The authors summarize the results of the inoculation of blood of early cases as follows: ". . . at least 50 per cent of the animals react in a characteristic manner. After a variable incubation period of not less than five days there is a more or less marked rise in temperature with or without catarrhal symptoms referable to the respiratory passages, such as sneezing and cough, and with or without an exanthem."

In the subinoculation of the virus in monkeys, the maximum incubation period was twenty-one days. Such irregularity complicates the interpretation of the data and increases considerably the difficulties of practical work. Unfortunately many details of the work are not available at present. In the majority of instances, the temperatures of the inoculated animal are not stated, since the com-

plete report of the work has not yet appeared. The character of the exanthem was extremely variable. Sometimes only an erythematous blush was noted. Frequently the rash was copper-colored from the beginning. Occasionally discrete pink macules and papules were observed which disappeared on pressure and were followed by a branny desquamation. These rashes occurred at very irregular intervals after inoculation; they developed most commonly on the face and chest but appeared sometimes on the thighs and abdomen. Rhinitis, coryza, and malaise were sometimes noted but these were not striking symptoms. No observations are recorded concerning leucocyte counts or examinations for Koplik spots.

Several strains were subinoculated from monkey to monkey. One in particular was passed rapidly through a series of 6 monkeys in forty-four days, but no evidence was noted of any alteration in its virulence. Experiments were also conducted to determine the infectivity of the blood for monkeys after infiltration, and after exposure to unfavorable conditions. Four specimens of blood were passed through a Berkefeld filter. Negative results were obtained with the first three; with the fourth specimen, 1 of 2 animals developed an exanthem twenty-one days after inoculation. Subinoculation of blood from this animal produced a slight febrile reaction in 1 of 2 monkeys. The authors conclude that the virus of measles is capable of passing through a Berkefeld filter.

Additional experiments were made concerning the effect of drying, heating, freezing, and of age upon the virus. They draw the following conclusions: "The virus in measles blood may resist desiccation for twenty-five and one-half hours, lose its infectivity after fifteen minutes at 55°C., resist freezing for twenty-five hours, and possibly retain some infectivity after twenty-four hours at 15°C."

Anderson and Goldberger also inoculated monkeys with mucous secretions of measles patients. Two monkeys inoculated on the mucous membrane with material taken twenty-five hours after the rash appeared, developed no symptoms. Subsequent work was carried out by subcutaneous injection of secretions. The contaminating bacteria produced a prompt rise in temperature and a local abscess. The latter was usually incised and some drop in temperature usually occurred. In some animals the temperature subsequently

rose again with or without the development of a rash. There were 5 experiments in which secretions were taken not later than twenty-six hours after the first appearance of the exanthem. In 4 instances the results were negative or doubtful. Secretions were obtained from 1 patient at the beginning of the rash and again twenty-four hours later. Successful inoculation of monkeys was reported with both specimens.

Hektoen and Eggers (10) supplied data more especially concerning the leucocyte counts in monkeys inoculated with measles blood. They report a more or less definite initial leucocytosis followed by a leucopenia of variable degree involving principally the neutrophils and resulting in a relative increase in the lymphocytes. In control animals injected with normal blood they noted either no change or else a slight transitory leucopenia. Two monkeys received measles blood obtained during the first twenty-four hours of the rash. One of these, on the twelfth and thirteenth days after inoculation showed signs of malaise, but there was no rash and no respiratory complications. The other developed evidence of malaise on the twelfth day; a faint masculo-papular rash appeared about the eyes and forehead on the fifteenth day, and a similar rash developed in both groins on the following day. These rashes disappeared after one to two days without any distinct desquamation. No Koplik spots were present. Subinoculation of monkeys was performed with blood taken late in the incubation period and no definite symptoms resulted.

The authors conclude that their results, when combined with those of Anderson and Goldberger, indicate that the *M. rhesus* is susceptible to a mild kind of measles.

Lucas and Prizer (11) described the occurrence of Koplik spots in monkeys. Two animals (*M. rhesus*) were injected with blood from a pre-eruptive case of measles. They report a leucopenia and the development of Koplik spots ten days after injection. On subinoculation into 2 other monkeys, spots, which were interpreted as Koplik spots, appeared in one, after ten days. The duration of these spots is not stated. The 2 animals injected with measles blood from man showed a transient erythema but no rash. No febrile reactions developed. The interpretation of these results is difficult because of an intercurrent infection of unknown etiology which killed several

control monkeys and also some of the inoculated ones about two weeks after their injection with measles blood.

Nicolle and Conseil (12) in 1911, reported confirmation of the work of Anderson and Goldberger. One monkey (*M. sinicus*) was injected with blood taken from a case of measles twenty-four hours before the rash appeared. The animal developed no symptoms except very slight malaise and a rather transient rise in temperature, most noticeable on the eleventh and twelfth days of the incubation period. Blood taken on the eleventh day was injected into a very young monkey (*M. sinicus*) but the animal remained entirely normal. The authors conclude that they have confirmed the work of Anderson and Goldberger.

In 1920, Nicolle and Conseil reported very briefly the results of some experiments conducted in 1913, concerning the transfer of measles from a child to monkeys (*M. sinicus*), re-inoculated successfully into a child, and again in monkeys. No symptoms other than a febrile reaction were observed in the monkeys; the temperatures are given for only a short portion of the incubation period. It is, therefore, inadvisable to draw any conclusions without knowing the normal temperature for these animals. As regards the child injected with blood from a monkey, there is no description of the symptoms, such as the respiratory involvement, Koplik spots, leucopenia, or glandular enlargement. There is no description of the rash, nor any reference to subsequent desquamation. It is certainly very important to know whether the course of the disease resembled the spontaneous infections, or whether some of the modifications occurred which were noted by Hektoen. This information is particularly desirable since there is no description of the precautions which were taken to avoid contact infection with measles.

Tunncliffe (13) inoculated one animal (*M. rhesus*) with blood from a measles patient taken at the end of the first twenty-four hours of the rash. There was no definite febrile reaction. The temperature at the time of inoculation was 104°F. It rose from 102.6°F. on the seventh day to 103.5°F. on the eighth day and then fell slightly. Tunncliffe considered that this rise may have been caused by the virus of measles. A protracted leucopenia developed, the count remaining relatively low, for fifteen days, a period which is much

longer than other observers have recorded in monkeys inoculated with measles; it is also much in excess of the duration of the leucopenia occurring in human cases. There was neither rash nor Koplik spots, nor other indication of measles.

Jurgelunas (14) endeavored to produce measles in monkeys by inoculation of blood, of mucous secretions, and by exposure of animals in a measles ward. He concludes that his results were negative.

One monkey (*Pavian*) was injected with defibrinated blood from a patient showing Koplik spots at that time; the rash appeared on the following day. Ten days after injection, the animal developed small rose colored spots over the abdomen. There was no rise in temperature. Death occurred on the following day. The autopsy failed to reveal the cause of death. The liver and spleen were enlarged. Cultures from the blood and various organs showed no growth. Jurgelunas considers that the rash did not conform to the exanthem of measles and that measles was not the cause of death in this animal. He injected one other monkey (*M. cynomolgus*) with the blood from an active case of measles, the specimen being taken during the first day of the rash. No symptoms developed. A third monkey injected with blood showed no symptoms, but it should be noted that the specimen was not taken till the second day of the rash.

Two monkeys were exposed to natural infection in a measles ward, being five days among acute cases and two days with convalescent patients. Neither developed any symptoms of measles; one, however, died of an acute streptococcus peritonitis two weeks after the last exposure in the ward.

Several experiments were conducted with mucous secretions, all of which were negative. One animal (*M. cynomolgus*) was injected subcutaneously with specimens taken on the day preceding the appearance of the rash. In another (*M. cynomolgus*), the mucous membranes of the mouth were rubbed with secretions from a patient showing Koplik spots but no exanthem. Another monkey (*M. rhesus*) was inoculated in the same way with specimens taken during the first day of the eruption. Lastly, the secretions from another case taken during the first day of the rash were rubbed into the scarified mucous membrane of the mouth of a *M. rhesus*.

Jurgelunas made no comments concerning leucocyte counts and Koplik spots, relying apparently on the temperature and an exanthem for indications of an infection.

Blake and Trask (15) have reported the successful infection of monkeys (*M. rhesus*). Ten monkeys were inoculated with the mucous secretions of early cases and 8 are regarded as having developed symptoms of measles. The authors confirm the occurrence of a rash, the febrile reaction and the malaise noted by Anderson and Goldberger, the leucopenia first noted by Hektoen and Eggers and occasionally found Koplik spots as reported by Lucas and Prizer. Many of their animals developed more or less conjunctivitis but none showed any rhinitis nor bronchitis. The filterability of the virus of measles was also confirmed. In 2 instances, mucous secretions of patients were passed through a Berkefeld N filter. The filtrate upon injection into monkeys, produced an exanthem and an enanthem but no fever developed.

The evidence of leucopenia as recorded in the charts is not particularly constant nor striking. However, the authors state that they do not regard the temperature and leucocyte counts as evidence of successful inoculation, but merely as additional data.

The characteristic enanthem in the monkey as noted by Blake and Trask consisted usually of a bright erythematous discrete or granular rash occurring most commonly on the labial mucous membrane and the gums. In one instance whitish lesions occurred resembling the Koplik spots of human cases. Histologically the cellular reaction of the enanthem and exanthem occurring in monkeys conformed to the description of the human lesions as given by Mallory and Medlar. Apparently no examinations were made for the Gram-positive coccoid bodies found by Mallory and Medlar in measles. These histological studies would be considerably strengthened in case the picture of these lesions proved to differ sharply from that of the spontaneous maculopapular rashes which often occur in monkeys.

The authors stress emphatically the very close resemblance of experimental measles in monkeys as compared with the human disease. The two differ significantly, in their opinion, only in the inconstant febrile reaction and the absence of rhinitis and bronchitis. To this I would add the usual absence of typical Koplik spots in monkeys and the inconstancy of a definite leucopenia.

The usual immunity tests were carried out, employing 6 monkeys which had shown a reaction to the virus of measles and 2 control monkeys. The 2 controls developed symptoms but the 6 which had previously reacted remained negative. The data concerning the temperature and leucocyte counts are not given.

Subinoculations from monkey to monkey were carried out, using either blood or the ground skin and mucous membrane of inoculated monkeys. The authors consider that the early transfers gave successful infections but that after repeated passage (8 to 12 transfers) a strain eventually dies out. In the inoculations made directly from patients and also in the subpassages, no febrile reactions developed except in those animals injected with contaminated material. In the course of the subinoculations, whitish areas resembling Koplik spots were noted in the enantheams which developed in 2 of 12 or more animals.

Four monkeys were injected intravenously with blood and all of these developed conjunctivitis. This result in a rather refractory species stand out in more or less contrast to the observation of Hektoen. It will be recalled that 1 of 2 volunteers, injected subcutaneously, escaped any signs of involvement of the mucous membranes and in the other only a mild conjunctivitis and some cough developed.

In the beginning of their work Blake and Trask applied the procedure of intratracheal injection for the inoculation of the virus in monkeys but they appear to have obtained satisfactory results with equal ease by rubbing infective material on the mucous membranes or by the injection of blood. Their experiments, however, were not designed to test the relative value of the various methods of inoculation.

Kawamura (16) took blood from a measles patient sixty hours before the appearance of the eruption and injected rather less than 1 cc. of blood into each of 3 monkeys (*M. fuscatus*). After an incubation period of eight or nine days, a fever, leucopenia, rash, conjunctivitis and rhinitis developed. Koplik spots were noted in 1 animal. Two successful subpassages were obtained by the injection of blood. Histologically, the rash in monkeys appears to have resembled both the cellular reaction seen in measles and also that of Japanese flood river fever.

In the course of their work on the inoculation of rabbits with measles, Nevin and Bittman (17) had occasion to inject 2 monkeys. One of these monkeys was injected intratracheally with mucous washings from an early case of measles; the other received blood of 2 patients taken early in the eruptive stage. The animals developed more or less leucopenia, an exanthem and an enanthem but no fever. In some later work these authors inoculated a third monkey with patient's blood under similar conditions and obtained a similar result.

In 1918 and 1919, Wentworth and the writer (18) carried out some experiments upon the inoculation of monkeys with measles. In a preliminary experiment 3 animals (*M. rhesus*) were used for blood injections. The first (A) was given 10 cc. of blood from a patient eighteen hours after the rash appeared. In transmitting typhus fever to monkeys, Ricketts and Wilder (19) recommend dilution of the blood. Accordingly this quantity of 10 cc. was diluted with 40 cc. of isotonic salt solution, defibrinated, and injected intraperitoneally. The animal remained well and there was no evidence of any rash or Koplik spots. The temperature and leucocyte count did not fluctuate beyond the normal limits.

A second animal (B) was injected with blood from a patient within six to twelve hours after the onset of the rash; 10 cc. were diluted with an equal volume of isotonic salt solution, defibrinated, and injected intraperitoneally. This animal was kept under observation for ten days before injection. During the early part of this period, a marked erythema with a few macules was present over the face and eyebrows. This rash practically disappeared during the first week of the incubation period, and then increased very slightly ten days after inoculation. Two months after the last injection it was more marked than at the beginning of the experiments. Otherwise, the findings in this animal were negative.

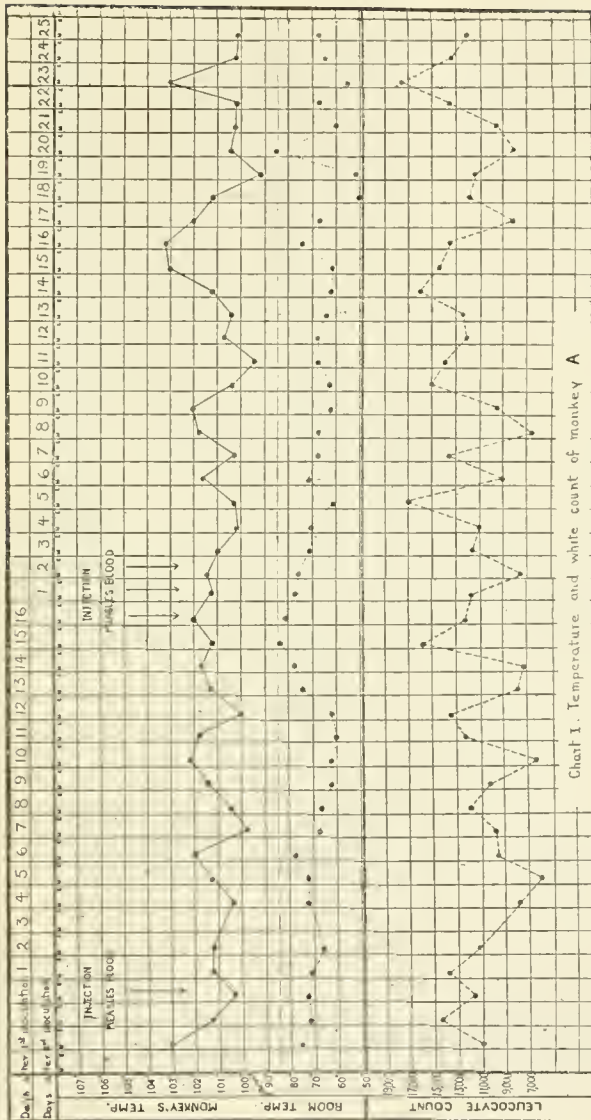
Very frequently, an animal which fails to respond to an injection with blood from a case of typhus fever may subsequently react typically to a similar injection. Accordingly these 2 animals (A and B) and a third young adult monkey (C) were given rather intensive injections of blood from measles patients. They were injected on three successive days with blood taken from 3 cases of measles

in the early stage of the exanthem. On the first day, blood was obtained from a patient four to five hours after the rash appeared; on the second and on the third day, from patients in each of whom the rash had started about twelve hours previously. The blood for these injections was either defibrinated or collected in sodium citrate.

There was no evidence of any reaction in these 3 monkeys. On the eleventh day after the first of the three injections, 3.5 cc. of blood was withdrawn from monkey C and injected subcutaneously in a susceptible volunteer. There was no change in his temperature or leucocyte count and no symptoms developed.

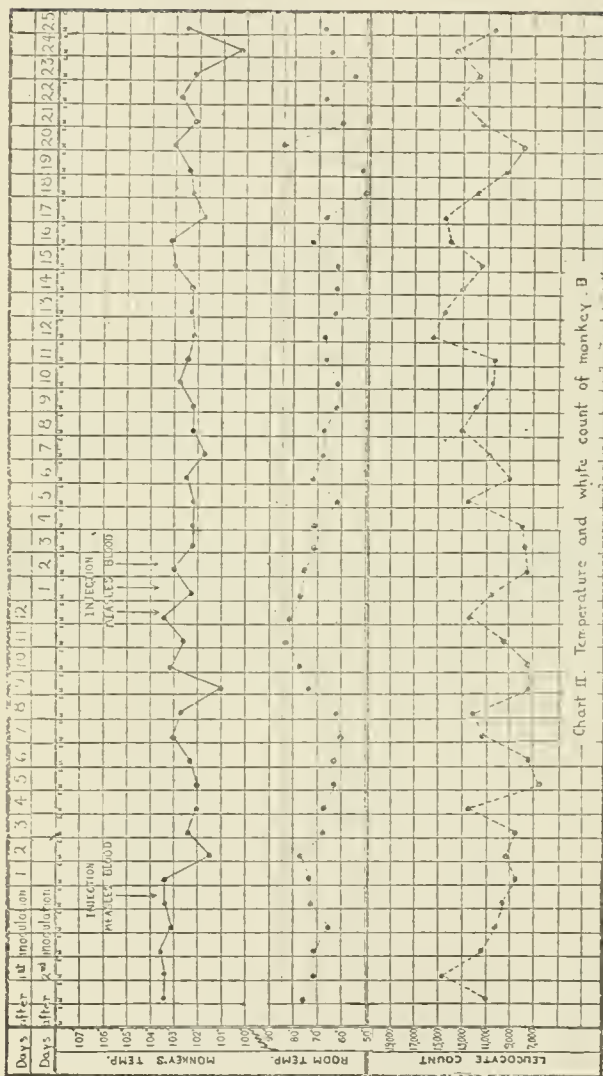
The leucocyte counts and the temperatures of these monkeys are given in charts I, II, and III. As an additional control, the room-temperature is also included since the body temperature of monkeys is sometimes influenced by this factor. These charts represent very clearly the disappointing type of reaction that may commonly be expected in monkeys even when inoculated under favorable conditions.

In a continuation of this work (20) some rather interesting results were obtained from an experiment in which portions of the same specimen of measles blood were injected simultaneously in 2 volunteers and in 2 normal monkeys (*M. rhesus*). As already described neither of the 2 men developed any symptoms; 1 of the 2 animals showed a suggestive reaction. In the interpretation of this result it must be recalled that 1 of these 2 volunteers gave exceptionally clear evidence of never having been exposed to measles. Blood was obtained from 2 patients for these injections, specimens being taken on 2 successive days. For the sake of convenience, the description of these cases will be repeated here. On the day of the first injection both patients were in the pre-eruptive stage. Pooled specimens of blood taken in citrate solution were injected at once. Each of the volunteers received the equivalent of 3 cc. of blood, the first portion being injected subcutaneously and the remainder intramuscularly. Each of the two monkeys received the equivalent of 2 cc. of blood, part of which was injected subcutaneously and the remainder intraperitoneally. One of the 2 measles patients developed a rash six hours after withdrawing the first specimen of blood. On the next day the patients were seen again; one was still in the pre-eruptive



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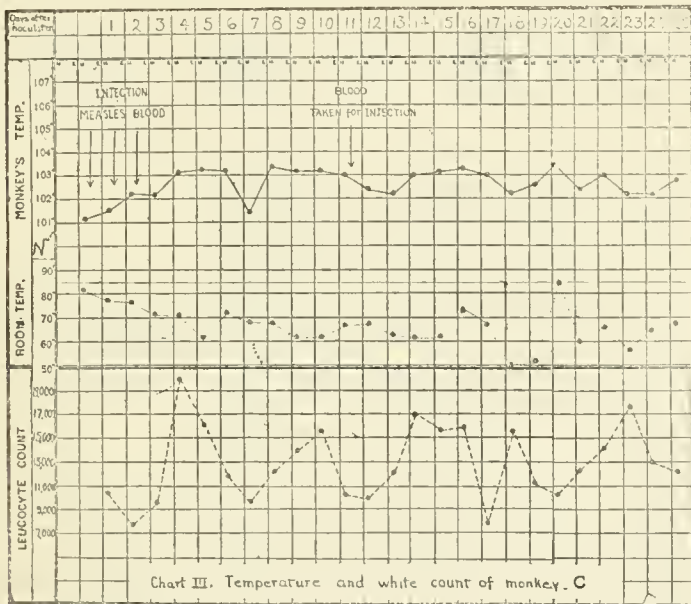
CHART I



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CHART II

stage but the rash appeared about six hours later. Blood was taken from both patients, the specimens were pooled and all of the injections were repeated as on the preceding day, employing the same quantities. The 2 monkeys varied somewhat in their reaction. One (D) showed a low leucocyte count on the ninth and again on the eleventh and twelfth days after injection. There was no febrile reaction, no respiratory nor constitutional symptoms, and no exanthem nor Koplik spots. The other animal (E) developed a leucopenia



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CHART III

beginning on the sixth day after his first injection, and persisting for three consecutive days. On the twelfth day, a faint rash developed over the face, neck, and uppermost part of the chest. This was principally a diffuse erythematous blush but there appeared around the eyes and nose discrete red macules 1 to 2 mm. in diameter from which the color could be readily expressed. On the next day, the rash faded almost completely leaving behind only slightly pigmented areas. These disappeared on the following day, and they were not followed by desquamation. On the first day of the rash a moderate

degree of malaise was noted. These symptoms were not accompanied by any febrile disturbance. There was no rhinitis and no Koplik spots were found at any time. On the fifteenth day after injection a well marked pneumonia developed.

On the seventh day of the incubation period, when the leucopenia appeared, 3 cc. of blood were withdrawn and injected subcutaneously

TABLE 1

DAYS AFTER FIRST INOCU- LATION	RHESUS D			RHESUS E			ADDITIONAL OBSERVATIONS ON RHESUS E
	Temperature		White count per cubic milli- meter	Temperature		White count per cubic milli- meter	
	a.m.	p.m.		a.m.	p.m.		
	°F.	°F.		°F.	°F.		
1		101.2	18,900		101.8	13,900	
2	99.0	102.0	11,100	100.4	102.2	10,700	
3	99.4	101.8	10,100	101.8	101.0	18,500	
4	101.0	102.4	11,800	102.0	102.0	10,000	
5	101.2	102.0	12,500	100.6	102.0	11,800	
6	100.8	102.2	14,900	102.2	102.8	5,500	
7	101.6	101.8	8,300	102.4	102.0	5,200	Bled for inoculation of volunteer
8	100.8	101.2	9,500	101.2	101.8	6,400	
9	100.8	102.4	4,700	101.6	102.0	7,900	
10	101.6		7,400	101.2		9,900	
11	101.4	101.8	4,400	102.8	103.6	11,110	
			4,900				
12	101.2	102.6	5,700	101.0	101.4	12,600	Slight rash
13	101.6	102.9	7,800	101.0	102.0	8,200	Slight rash
14	100.9	102.2	9,200	100.0	101.0	6,400	
15	101.4	102.0	8,900	100.0	100.0	6,000	Early signs of pneumonia
16	100.6	101.0	8,300	99.4	99.8	6,600	Definite pneumonia
17	100.6	102.2	17,300	98.8	100.6	18,400	Critically ill
18	102.0	102.2	13,900	101.0		9,400	Critically ill
20	101.2		14,500	100.8		44,000	Marked improvement by crisis

and intramuscularly into a volunteer. No local nor general symptoms developed.

The record of the temperatures and white counts for the two monkeys are given in Table 1.

The rash and leucopenia developing in this second monkey, unaccompanied by rhinitis, fever or Koplik spots are difficult of interpre-

tation. The absence of any symptoms following the corresponding injection of measles blood in man constitutes strong evidence against ascribing the reaction in the monkey to the virus of measles. This is especially true in view of the direct concrete evidence of susceptibility in one of these volunteers. On the other hand, the reader may on purely general grounds feel skeptical about the susceptibility to measles of any adult. It must also be remembered that each of the monkeys received some measles blood intraperitoneally. In view of the ultimate results, the experiment is faulty in this respect; for it is theoretically possible that a refractory animal might be overwhelmed by an intraperitoneal injection although a susceptible host escaped infection after subcutaneous and intramuscular injection.

It is noteworthy that the first of these 2 monkeys remained free from any characteristic reaction notwithstanding the intensive injection of extremely favorable material. This result might be taken as an illustration of Anderson and Goldberger's view that many individual animals are altogether refractory.

My own experience with the inoculation of monkeys with mucous secretions has given only negative results. I have endeavored to infect 2 monkeys (*M. syriacus*) by inoculation with secretions taken four days and one day before the patient's rash developed. Swabs moistened with the conjunctival secretions of the patient were rubbed over the conjunctivae and nasal and pharyngeal mucous membrane of the monkeys. Similarly swabs from the nasal and pharyngeal mucous membrane of the patient were thoroughly rubbed over the corresponding mucous membranes of the animals. Neither monkey developed any fever or leucopenia. There was no rash nor Koplik spots, no inflammation of the mucous membranes, and no malaise. Two additional monkeys (*M. rhesus*) were inoculated with secretions from a measles case taken two hours after the first appearance of the rash. The inoculations were made in the same manner as for the syriacus monkeys. In addition, scarified areas of the mucous membrane of the monkey's mouths were rubbed with swabs from the conjunctival and nasal mucous membrane of the patient.

For the sake of convenience, table 2 has been prepared showing the general results obtained by various investigators upon injecting monkeys with the blood of measles patients. This outline covers

only those experiments which were designed to determine whether the monkey is susceptible to measles. It does not include the records of those injections in which the patient's blood was subjected to various procedures such as filtration or aging, for the purpose of studying the properties of the virus. Reports based upon the injection of a single animal are also omitted.

The results concerning the inoculation of the mucous secretions as obtained by various investigators are given in the table 3. The negative experiments conducted with late cases are not included.

TABLE 2
Inoculation of monkeys with blood of early cases of measles

EXPERIMENTAL RESULTS	ANDERSON AND GOLDBERGER	HEXTON AND EGGES	LUCAS AND PRIZER	NICOLLE AND CONSEIL	JURGELUNAS	KAWAMURA	NEVIN AND BITTMAN	SELLARDS AND WENTWORTH
Incubation period, days.....	5-11	12	10	11	---	8-9	4	---
Fever.....	+	+	0	+	0	+	0	0
Leucopenia.....	---	+	+	---	?	+	+	+
Exanthem.....	+	+	?	0	0	+	+	?
Enanthem.....	---	0	+	---	---	---	+	0
Conjunctival or respiratory signs.....	+	0	?	---	---	+	+	0
Malaise.....	+	+	---	?	---	+	+	?
Subinoculation in monkeys.....	+	?	?	+	---	+	---	---
Re-inoculation in man.....	---	---	---	+	---	---	---	0
Number of animals inoculated.....	7+	2	2	6	2	3	3	5
Number showing symptoms.....	4+	2	2	5	1?	3	3	1

0, none; ---, no observations; +, present; ?, irregular or doubtful.

Localized lesions. In the attempts to reproduce measles in animals, practically all of the attention has been directed toward obtaining a systemic infection. In this connection, a consideration of smallpox is instructive. The virus of smallpox certainly gains access to the circulating blood at some periods of the infection. Yet the experimental transfer of the disease by the injection of blood has not been conclusively demonstrated. However, subinoculation from skin lesions in man to the skin of lower animals, readily produces a local lesion but a generalized infection typical of the spontaneous disease has not been obtained. In the course of some unpublished work, Bigelow and the writer carried out an analogous procedure in measles.

Early skin lesions and Koplik spots were excised from patients and implanted in the skin and mucous membrane of monkeys. Several of the results were entirely negative but some were very suggestive. In one instance in particular, an implant of skin lesion into the skin was followed after two weeks by the development of bright pink papules in an area approximately 5 cm. in diameter surrounding the implanted tissue. These papules faded gradually in the course of three days and were followed by pigmentation and desquamation.

TABLE 3
Inoculation of monkeys with mucous secretions of patients

EXPERIMENTAL RESULTS	ANDERSON AND GOLDBERGER		JURGELUNAS		SELLARDS	BLAKE AND TRASK
	Swabbing mucous membrane	Subcutaneous injection	Swabbing or scarifying mucous membrane	Subcutaneous injection	Swabbing or scarifying mucous membrane	Swabbing mucous membrane or intratracheal injection
Incubation period, days.....	—	8 and 9	—	—	—	6-10
Fever.....	0	+	0	0	0	+
Leucopenia.....	—	—	—	—	0	+
Exanthem.....	0	+	0	?	0	+
Enanthem.....	—	—	—	—	0	+
Conjunctival or respiratory signs...	0	+	0	0	0	+
Malaise.....	0	+	0	—	0	+
Subinoculation in monkeys.....	—	+	—	—	—	+
Number of animals inoculated.....	2	6	3	1	4	10
Number of negative or doubtful reactions.....	2	2	3	1	4	2

0, none; —, no observations; +, present; ?, irregular or doubtful.

Normal human skin implanted in control monkeys was gradually absorbed without producing any eruption.

In seeking for a method of active immunization against measles, Blake and Trask (21) report the development of a localized lesion in monkeys by the intramuscular injection of an attenuated virus. It is well to recall that Hektoen, injecting the virus of measles subcutaneously in man, observed no trace of any local reaction.

Discussion of the reaction in monkeys. There is certainly, at present, no exact proof of the susceptibility of monkeys to measles. The work of Nicolle and Conseil suggests that the virus of measles is

conserved in monkeys and may produce a mild febrile reaction. It will be recalled that these observers noted the development of measles in a child which they inoculated with blood from a monkey showing a mild reaction some days after being injected with virus. This observation cannot be accepted as final without knowing the precautions which were taken against accidental infection.

In contrast to experimental typhus fever, and spotted fever, no criteria have as yet been established by which experimental measles can be recognized definitely and unmistakably. Spotted fever in the guinea pig runs a fatal course with a characteristic pathology. A guinea pig inoculated with typhus fever develops only a moderate febrile reaction and no symptoms appear which would suggest the human disease. However, microscopic lesions occur with great regularity in the brain; the histologic picture of these corresponds to those found in human cases and furnishes an accurate method of recognizing the experimental disease. The rashes sometimes occurring in monkeys inoculated with measles agree fairly well in the histologic picture with the skin lesions of human cases. This histologic picture, however, is not pathognomonic of the disease.

Several species of the genus *macacus* have been utilized more or less extensively, namely, *rhesus*, *cynomolgus*, *sinicus*, and *fuscatus*. Thorough comparative studies are not available but there is no indication that the characteristics of the reaction to the virus is dependent on the type of monkey employed.

The mass of evidence though conflicting in many respects, suggests on the whole that occasional individual monkeys (genus *macacus*) may show mild reactions of variable character when injected with the virus of measles. Are these signs and symptoms sufficiently frequent and definite to constitute a reliable method for the experimental study of measles? If, for example, only 1 animal in 4 or 5 gives a definite dependable reaction, then the method soon becomes unwieldy and impractical for any extensive studies.

Observations in monkeys may be controlled in a measure by the "immunity test." But this test loses much of its significance since, even after intensive inoculation with ideal material, a considerable proportion of animals remain essentially free from symptoms.

The acceptability of these delicate reactions occurring in monkeys as a reliable method for the study of measles resolves itself ultimately into a question of the standards which the individual investigator considers essential. To me, they are not satisfactory. Personally, I am not willing to accept as established the various characteristics of the virus of measles as worked out in this way. Thus the important conclusion that the virus is filterable rests primarily upon more or less vague results obtained in three monkeys. I prefer to consider the filterability of the virus as an entirely open question.

Inoculation of rabbits and guinea-pigs. In the past two years, a few attempts have been made to simplify the study of measles by the substitution of rabbits or guinea-pigs for monkeys in experimental work. Nevin and Bittman took blood from 6 cases of measles, two to four days "after the onset of the disease." Six rabbits were inoculated intravenously and all gave evidence of a reaction. There was no characteristic fever nor leucopenia. The animals were shaved before inoculation. The redness caused by shaving became more intense in those receiving blood and subsequently desquamation occurred. In the control series, the redness after shaving faded without desquamation. Subinoculations of blood were made from rabbit to rabbit and 9 of 11 animals reacted. One strain, after five passages in rabbits, was inoculated into a monkey, *M. rhesus*. A somewhat suggestive leucopenia developed on the third day; the following day two spots somewhat resembling Koplik spots appeared on the labial mucous membrane; then a maculo-papular rash appeared on the face and later a red granular rash on the mucosa of the lips. The exanthem was followed by a marked desquamation. Subsequently, this animal showed no reaction to an intratracheal injection of 10 cc. of mucous washings from a patient with measles. The authors consider that this monkey developed typical measles as a result of the injection of blood from the inoculated rabbits and was, therefore, immune to the injection of secretions from a patient with measles. To me it seems equally possible that the rash developing in the monkey after an inoculation of rabbit's blood was not necessarily produced by the virus of measles; also the failure to react to a test injection of mucous secretions may have been nothing more than the corresponding failures which have been noted from time to time in normal monkeys.

In some later work, Nevin and Bittman passed a strain of measles virus through three rabbits and then through a series of threemonkeys in order to eliminate as far as possible any question of rashes due to a foreign protein. Leucopenia, Koplik spots, an enanthem and an exanthem were noted in all of the monkeys after injection with blood from inoculated rabbits. Some of the rabbits, in addition to an erythema, developed a generalized maculo-papular rash followed by pigmentation and extensive desquamation. Koplik spots and enanthems were also noted. The authors conclude that the virus of measles "survives passage in rabbits."

Simultaneous with the studies of Nevin and Bittman on the blood of measles cases, Grund (22) working in the same laboratory collected mucous secretions from these same patients and injected rabbits intratracheally. Of 23 animals a rather large number proved refractory. No definite febrile reaction or leucopenia occurred. In 1 or 2, a maculopapular eruption developed and in 10 or 11 an erythema occurred. Sub-passages in rabbits gave somewhat more encouraging results. Immunity tests on convalescent animals proved rather "contradictory." Grund concludes that no one individual animal gives a typical picture of measles but that the series, taken as a whole, encourages the belief that rabbits are susceptible to the virus of measles.

Duval and D'Aunoy (23) conclude that rabbits are susceptible to measles developing a specific reaction which they regard as analogous in all essential features to the human disease. They consider "only temperatures of 102°F. or over as pyrexia" and regard white counts under 9000 cells as evidence of leucopenia. After the intravenous injection of patient's blood in rabbits, they noted the development of coryza, conjunctival injection, and enanthem similar to Koplik spots and in 40 per cent of the animals an exanthem appeared. A number of rabbits developed an acute hemorrhagic nephritis.

After several subpassages in rabbits, a very remarkable phenomenon was noted by Duval and D'Aunoy. They report a striking increase in virulence and conclude that a number of animals died undoubtedly from the direct effect of the virus of measles and not on account of intercurrent infection. This finding would require extensive confirmation and elaborate control in order to eliminate the possibility of epizootic disease.

The susceptibility of guinea-pigs to measles was studied also by Duval and D'Aunoy. They conclude that the guinea-pig reacts specifically to the virus of measles showing a definite and constant rise of temperature with a coincident fall in the total number of leucocytes after an incubation period of nine to twelve days.

Several large series of experiments were conducted but unfortunately some of the essentials of these data do not appear in the report. Those portions of the data concerning the temperature and leucocyte count which the authors present are not sufficiently complete to permit a logical conclusion. The situation in brief is as follows: The temperatures and leucocyte counts of 30 normal guinea-pigs were taken for thirty-one days and the daily average result of this series is recorded. In a similar manner, 15 guinea-pigs were injected with normal human blood and the daily average temperature and leucocyte count is recorded. Finally, the blood from 7 cases of measles was injected into guinea-pigs. In each experiment of this series, 6 animals were used, 4 for blood from measles patients and 2 for controls. Thus 28 pigs received measles blood and 20 of these showed evidence of reaction. However, only two charts are given of temperature and leucocyte counts and it is entirely impossible to determine whether these charts represent the data of a single animal or the composite data of more than one. Since some of these animals were sacrificed, the curve is not a composite of the entire group. Obviously the chart of a single experiment or of an entirely unknown number of animals cannot be compared with the composite chart of 30 control animals, studied one or two months previously. The 14 control animals inoculated simultaneously with those receiving measles blood showed no reaction but no data are given. It would appear that any temperature above 102° was regarded as abnormal. In passage experiments from guinea-pig to guinea-pig, the virus increased in virulence even to the extent of killing "a number" of the animals. Acute hemorrhagic nephritis is reported as a constant finding but unfortunately the number of animals examined is not indicated.

Tunncliffe and Moody (24) injected 9 rabbits intratracheally with the virus of measles using presumably mucous secretions. Good rashes were observed in 8 of these animals but no other definite

symptoms developed. None of 15 control rabbits showed any rashes similar to those produced by the virus of measles. Two guinea-pigs were inoculated intratracheally with the virus of measles, the results suggesting a rise in temperature and in one instance a leucopenia.

Kawamura inoculated monkeys with the blood of measles patients and subinoculated from the monkeys into guinea-pigs and rabbits with entirely negative results.

Nicolle and Conseil inoculated rabbits and guinea-pigs and conclude that these animals are not susceptible to measles.

In conclusion, it would seem clear that the symptoms in rabbits appear even less definite than those described in the monkey and the evidence that the virus survives in rabbits rests, in a large measure, on the re-inoculation from rabbits to monkeys. Acceptance of the susceptibility of rabbits and guinea-pigs to measles, or even the survival of the virus in these animals, is not warranted on the evidence which has been submitted.

BACTERIOLOGICAL INVESTIGATIONS

Cultural work on measles has been restricted, for the most part, to the aerobic bacteria. The types of media and the methods which have been employed have not differed strikingly from the standard routine bacteriological procedures. The materials usually selected for examination have been the secretions of the conjunctival and upper respiratory mucous membrane, the circulating blood and the lesions of the skin. From these sources, many representatives of the common types of bacteria have been isolated, none of which occur constantly or exclusively in measles. The organisms which have been described are not very remarkable. The earlier observations have been summarized by Hektoen (25).

B. influenzae. Of the various bacilli observed in measles, *B. influenzae* of Pfeiffer has attracted the most attention. It was first isolated from uncomplicated cases of measles during the active stage of the disease by Giarre and Picchi (26) in 1900. Subsequent observers have found that this bacillus appears with considerable frequency in measles. It occurs rather abundantly in the mucous membrane of the conjunctivae and upper respiratory tract, and in the lungs in cases complicated by pneumonia, but it has not been found in the blood nor in the skin lesions.

Several weeks after the subsidence of the first wave of the pandemic of influenza in 1918, the occurrence of *B. influenzae* in cases of measles was investigated at Camp Devens, Massachusetts, by Lieutenant Sturm and the writer (27). As this was one of the camps in which the Pfeiffer bacillus was prevalent during the epidemic of influenza, it is obvious that this organism may have been rather widely distributed at this time. We recovered an organism indistinguishable from *B. influenzae* from the sputum in 80 per cent of a series of 31 consecutive cases of measles. Moreover in three-fourths of these patients, the bacillus disappeared with the subsidence of the acute symptoms. In a group of 7 control individuals, some of whom had had influenza a few weeks previously, repeated examinations failed to show the presence of the Pfeiffer bacillus. The strains obtained from measles cases were compared carefully with similar strains isolated at autopsy from the lungs of patients dying from complications of influenza. No significant differences were found either in morphology, staining properties, cultural characteristics, immunity reactions, nor in the behavior of these strains to freezing, drying, and the action of bile and sodium hydroxide. Two monkeys were inoculated with strains obtained from measles patients. One remained well but the other, after six days, developed pronounced malaise with cyanosis and a fall in the leucocyte count but without a rise in temperature. Two human volunteers who, as far as could be determined, had had neither measles nor influenza, were inoculated by rubbing strains from the cases of measles on the mucous membrane of the conjunctivae, the nose, mouth, and throat. No symptoms developed and no change occurred in the temperature or white count. Moreover the inoculated organisms could not be recovered in subsequent cultures. Under natural conditions the Pfeiffer bacillus frequently establishes itself upon a mucous membrane. Therefore, the failure to produce symptoms would have had much more significance if successful colonization had been obtained. Mallory and Medlar found *B. influenzae* frequently in measles cases in 1916 and only rarely in 1917.

Various interpretations have been suggested concerning the occurrence of the influenza bacillus in measles. Several of the early observers considered that the presence of this organism justified the diagnosis of a complicating influenza. This conclusion is not war-

ranted since, as a rule, the cases of measles harboring the bacillus do not show clinical evidence of influenza. A few observers, notably Giarre and Picchi, consider the possibility that a Pfeiffer-like organism may be the etiological agent in measles. In order to give favorable consideration to this view it is of vital importance to establish differences between the strains of the Pfeiffer bacillus occurring in measles and in influenza. Such differences have not been forthcoming. In their absence, the weight of evidence indicates strongly that the Pfeiffer bacillus is merely a secondary invader which multiplies more readily during the period of inflammation produced by the virus of measles. Indeed, the acceptance of the Pfeiffer bacillus as the specific cause of influenza is somewhat compromised by the frequency with which this organism appears in uncomplicated cases of measles.

Cocci. Many observers have described micrococci, very commonly in the mucous secretions, and much less often in the blood stream. The most recent observations are those of Tunncliffe (28) who isolated, by anaerobic methods, a micrococcus from the blood during the pre-eruptive and eruptive stages of the disease. In the latter stage, a very considerable variety of other organisms was also obtained. The micrococcus isolated from the blood, developed aerobically on subinoculation, producing green pigment on blood agar plates. A similar coccus was also found in the secretions from the mucous membrane. In a subsequent study, Tunncliffe confirmed the occurrence of this micrococcus in the mucous secretions but no report was made in regard to blood cultures.

In a recent paper Tunncliffe and Moody (24) describe the effect on animals of this micrococcus. Upon intratracheal injection in rhesus monkeys, a leucopenia developed but no rise in temperature occurred. Occasional small red papules appeared in the skin, the histological picture of these being consistent with the diagnosis of measles.

The cultures of this green producing coccus were also found to be pathogenic for rabbits, dogs, mice, rats and guinea-pigs. In the guinea-pig, a rise in temperature accompanied by a fall in the leucocyte count was noted, but no rash developed. In rabbits (22 in all) no definite fever nor leucopenia occurred but 42 per cent developed spots which were interpreted as Koplik spots and in 87 per cent an

exanthem was observed. Rats and young dogs injected with this micrococcus became ill but recovered. It is noteworthy that these animals are ordinarily regarded as being entirely refractory to measles.

Tunncliffe and Moody cautiously, and it seems to me wisely, refrain from concluding definitely that this green producing coccus is the etiologic agent of measles. In this connection it is interesting to recall that other micrococci are capable of producing rashes, notably the meningococcus and probably some of the streptococci as recently reported by Dick and Dick (29).

Wentworth and the writer, as well as Mallory and Medlar, were unable to demonstrate any micrococci in cultures from the blood of measles patients.

Diphtheroids. The diphtheroid bacilli represent one additional group of organisms which has been found more or less frequently in measles. Bacteria of this type appear to be almost omnipresent as a part of the normal flora of the body tissues. Observations of some interest were made by Ciaccio (30). In the autopsy of 8 cases of measles an organism, which was apparently a small diphtheroid, was found rather widely distributed in the lymph glands and in various organs, but never in the skin lesions. Mallory and Medlar in the cultural examination of the nasal and nasopharyngeal secretions noted the presence of diphtheroid bacilli almost constantly.

Bigelow and the writer (31) have frequently obtained diphtheroids, often in pure culture, from the conjunctivae of measles patients. The original cultures grew feebly and slowly, but eventually subcultures on ordinary egg or Loeffler's medium gave abundant growth. Subsequently we found repeatedly that a small Gram-staining pleomorphic bacillus developed in blood cultures from measles patients. This organism was obtained in 25 of 31 cases. In control blood cultures in 24 instances, growth occurred in 5 cases. The organisms obtained from the controls resembled those from the measles cases in their morphology and staining reactions but differed in their fermentation tests rather markedly from the majority of those obtained from the measles cultures.

Three rhesus monkeys were inoculated with cultures from the measles cases. In 2, the symptoms were vague but in the third a suggestive cluster of macules and papules developed. The histological picture conformed to the description of the human lesions.

Neither the micrococcus described by Tunncliffe nor the bacillus cultivated by Bigelow and myself have been confirmed by independent observers. The simplest explanation for both of these microorganisms is that, during the extensive inflammation of the mucous membrane, some of the normal flora are swept into the blood stream. Such an explanation is, of course, not an easy one to demonstrate experimentally. However, all workers in problems of etiology must bear in mind the now numerous examples in other infectious diseases of the cultivation, apparently from the blood stream, of bacteria which are surely without direct etiologic relationship to the disease.

RÉSUMÉ

Of the acute exanthemata, measles is the most important cause of infant mortality. The disease does not *per se* produce fatal results but only through its complications; one attack confers marked immunity. There is, therefore, an excellent theoretical basis for the development of a method of active immunization, this being obviously the most desirable procedure for bringing the disease under control. Practically, the experimental problem of developing such a method of inoculation has proven to be very difficult of approach. The difficulty might be solved, of course, either by obtaining a suitable source of supply of the virus with the development of a process for its attenuation or more ideally by the isolation and cultivation of the causative microorganism.

The clinical features of measles indicate that one might reasonably expect to find the virus of measles in the circulating blood of a patient and that the injection of such blood into a susceptible individual might reproduce the disease more or less regularly and perhaps in a somewhat modified form. The symptoms of the disease, the mode of infection and the resulting immunity suggest that the causative agent is not a typical protozoan; it may not unlikely prove to be a member of the general group of bacteria or the bacteria-like microorganisms, either visible or ultramicroscopic in size.

Histology. Careful histologic examinations of the skin lesions and the Koplik spots have not revealed any definite microorganisms. Nevertheless, the causative organism is probably present in these

lesions, perhaps in very scanty numbers. Neither has any cellular reaction been described which is diagnostic of the disease, the principal characteristic being some proliferation in the tissues around the vessels, of the endothelial leucocytes, the latter often showing mitoses. There is no evidence of primary necrosis or acute exudation of polymorphonuclear leucocytes such as the ordinary micrococci produce.

Bacteriology. Cultures of the inflamed mucous membranes have shown for the most part only the flora commonly occurring in the upper respiratory tract such as the cocci, the diphtheroids and frequently the influenza bacillus. A number of microorganisms have been found from time to time in cultures of the blood; two are worthy of mention; namely, the micrococcus obtained by Tunncliffe and a Gram-positive pleomorphic bacillus reported by Bigelow and the writer. Each of these organisms when inoculated in monkeys produced maculopapular lesions, the histology of which was consistent with that of human measles. In my opinion, this finding is not sufficiently distinctive to justify one in placing confidence in either of these organisms as the etiologic agent.

Transmission to man. It has already been emphasized that the existence of the virus of measles in the circulating blood of a patient does not necessarily presuppose that the injection of such blood in a susceptible person would produce an infection. The most valuable and the one most definite experimental contribution to the study of measles was made by Hektoen when he produced measles artificially in 2 volunteers by the inoculation of blood from a patient. He demonstrated at the same time that the virus will survive in ascitic broth at 37°C. for at least twenty-four hours. The clinical symptoms in these volunteers differed in minor respects from the usually constant picture of the natural infection. Information is lacking concerning certain features such as Koplik spots and the leucocyte counts. Indeed it is not yet established in how far "measles inoculata" might vary from the spontaneous disease.

My own work on the inoculation of volunteers with blood of measles patients has given only negative results, indicating that the injection of a patient's blood will not regularly and constantly reproduce the disease in individuals who are apparently susceptible.

Susceptibility of monkeys. Experimentally, one of the most important factors in the study of measles is the question of the susceptibility of monkeys. Attempts have been made in two directions to establish proof of the susceptibility of monkeys to measles. Nicolle injected blood from a measles patient into a monkey and noted a mild febrile reaction. A child inoculated with blood from this animal developed measles. Unfortunately the precautions which were taken to prevent accidental infection are not described.

Blake and Trask found that the histologic picture of the skin rashes occurring in monkeys inoculated with measles corresponded to the histology of the lesions of human cases. This histologic picture is not pathognomonic. We have, therefore, no convincing proof of the susceptibility of monkeys.

Although my own attempts to infect monkeys have been disappointing, nevertheless it seems to me that the weight of evidence in the literature favors the conclusion that occasionally individual animals develop mild reactions when inoculated with the virus of measles. However I am not willing to place dependence on this method for studying the disease. Practically all observers agree that the symptoms are rather vague, many individual monkeys being entirely refractory. Variation occurs in this respect to a much greater degree for example than in the case of the experimental production of typhus fever. Moreover, experienced investigators report altogether conflicting results in the study of measles regarding such cardinal factors as the development of a skin rash and the occurrence of a febrile reaction. There is also marked variation concerning details such as the incubation period, the presence of Koplik spots, of leucopenia, rhinitis and malaise. Anyone contemplating the study of measles in monkeys will find that very naturally no uniform technique has as yet been evolved. In the choice of material for inoculation, equally good results have been reported by the use of either blood or mucous secretions. Three modes of procedure have been employed for the inoculation of mucous secretions; namely, (1) swabbing the mucous membranes with or without preliminary scarification, (2) subcutaneous injection, and (3) intratracheal injection. No comparison of these methods has been attempted but theoretically the intratracheal injection when followed by regurgitation with coughing

and sneezing would give opportunity for a thorough inoculation of the mucous membranes.

Whatever the mode of inoculation, the chief difficulty arises in the interpretation of the reactions. Of the various findings reported in the monkey, there are three features of cardinal importance; namely, (1) fever, (2) leucopenia, and (3) rash, either of the skin or mucous membranes. These symptoms supposedly characteristic of experimental measles, are too mild to determine convincingly the etiologic relationship of suspected microorganisms isolated from patients.

This may seem to be an unhopeful view. On the contrary, it is merely suggested that attention should be directed toward a further study of the reactions in animals. It seems to me important to establish first of all an exact method of study rather than to increase the mass of data that has been founded on more or less doubtful methods.

Of the cardinal problems yet to be solved in measles we may mention: (1) the demonstration of the causative microorganism, (2) its cultivation, and (3) the infection of lower animals in such a manner as to provide a reliable and practical method for the recognition of the virus. By contrast with measles, let us consider a disease such as spotted fever, in which the causative organism is readily demonstrated microscopically in tissues and which produces in guinea-pigs a fatal infection with characteristic lesions. In any attempts at cultivating this organism, suspected cultures can be tested readily and conclusively. However, in measles, in working on any one of the three features just mentioned, it is necessary to contend with two unknown factors.

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ON THE PATHOGENESIS OF TETANY

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The literature concerning tetany is filled with somewhat disconnected observations of various symptoms and alterations of tissue which at first sight seem but little related. Such observations show that tetany is associated with rickets, or with pyloric obstruction, or with some infectious or renal disease. It follows destruction of the parathyroids sometimes accidentally brought about in operations on the thyroid; it can be made manifest when latent by certain intoxications or by lactation, although it is ameliorated by pregnancy. It can be produced by forced rapid respiration or by the intravenous injection of some substances such as alkaline bicarbonates and phosphates. It has as its most constant and easily recognized symptom the mechanical and electrical hyperexcitability of the nerves which may lead to tonic and clonic spasm of the muscles, but it may also be accompanied by changes in the teeth, eyes, skin, etc.

Indeed, one is led to believe that there may be several types of tetany differing widely in their etiology and in the mechanism of their production although the final changes in the blood which bring about the actual symptoms may be the same. Thus it must seem obvious that there can be little immediate relation between the extirpation of the parathyroid glands and a constriction of the pylorus, and that the changes in the blood through which they finally bring about similar symptoms must follow rather long converging paths until the ultimate change in the blood is the same in both. So, too, we might contrast the tetany of infants with that produced by prolonged forced respiration. The final status of the blood which causes hyperexcitability of the nerves may be the same but it has been reached by different processes. It seems even possible that we might deliberately produce that final status by various different ways of interfering with the com-

position of the blood, and we know positively that we can restore the animal to the normal state, temporarily at any rate, by reëstablishing the normal composition of this altered blood.

Observations upon the relations between tetany and psychoses, infections, intoxications and various disturbances of growth connected with deficiencies in one or other of the organs of internal secretion are still rather vague, as shown by Aschenheim in his recent careful review, but there are several types of tetany which stand out very distinctly and have been reasonably well studied. These are (1) the experimental or post-operative tetany resulting from extirpation or destruction of the parathyroid gland, (2) gastric tetany associated with long continued narrowing of the pyloric orifice, (3) tetany from forced respiration such as sometimes occurs in the hysterical or insane, and (4) infantile tetany or spasmophilia which accompanies rickets. There are doubtless others which occur spontaneously and still others which are of an artificial character produced by profound modifications of diet or by deliberately disturbing the constitution of the blood by massive injections of various substances such as bicarbonates or alkaline phosphates.

Experimental parathyreoprive tetany: The sequence of the most typical form of tetany upon the extirpation of the parathyroid glands is now perfectly recognized by everyone and most of the studies of experimental tetany have concerned this form. Its precise, and one might say, quantitative relation to the parathyroid glands is established although only after long dispute.

Post-operative tetany in man: Practically identical with this is the tetany which occurs after operations involving removal of large portions of the thyroid gland, especially in cases of carcinoma of the thyroid in which removal or destruction of the parathyroid glands may occur. A considerable literature has arisen recently with regard to precautions which may be observed in order to avoid actual removal or even the impairment of the circulation of these glands. But the literature is more especially concerned with efforts directed toward the prevention and cure of this post-operative tetany by the reimplantation of such parathyroid glands as are recognized after removal or by the transplantation of glands from other persons. Halsted's experimental studies gave an impetus to this effort by showing that

a parathyroid gland could be implanted in the same animal from which it was taken if a parathyroid insufficiency existed and that when established it would protect the animal from tetany even when all the other parathyroid glands were removed.

Von Eiselsberg, Flörcken and Fritsche, Roth and others have been successful in such transplantations in human beings and Borchers reviews this aspect of the subject up to 1921. Leschke thinks there may be a rachitic basis for post-operative tetany since the danger of its occurrence has increased with the increase of rickets since the war and the trauma of operation alone, without destruction of the parathyroids, is enough to cause it. Melchior, too, finds that since the war there is frequent alteration in the electrical excitability of the nerves and any operation may initiate tetany. This seems hardly comparable with post-operative tetany in previously normal people but rather the appearance of manifest tetany in persons in whom tetany was latent and in whom the operation is merely the incident which acts as a final stimulus to the over-excitable nervous system.

Gastric tetany in adults and children and in experimental animals differs profoundly in its origin from parathyroid tetany, inasmuch as the parathyroids are normal and with obstruction at the pylorus tetany develops in spite of the activity of the parathyroid glands. It appears from the work of MacCallum and associates, McCann, Hastings and Murray, and others to be due to a loss of hydrochloric acid through vomiting.

Tetany from forced respiration is quite analogous to this since carbonic acid is too rapidly removed from the body. Grant and Goldman, and others, have shown that this is followed by a decrease in the hydrogen ion concentration, an increase of the carbon dioxide combining power of the blood, alkaline urine, decreased excretion of ammonia and slight increase in the calcium of the serum.

Infantile tetany is certainly very different from the foregoing forms in its origin, inasmuch as it seems highly improbable that it depends upon destructive changes in the parathyroid glands. It is true that Yanase and Erdheim proposed the idea sometime ago that hemorrhage into the substance of the glands is responsible for the tetany but this has not been supported by others who find that hemorrhages are about as frequent in children that show no tetany and are often missing

in those that do. But in place of this suggestion there is no satisfactory explanation to offer. Much has been learned as to the metabolic disturbances but nothing as to the primary cause. The invariable association with rickets shows the character of this disturbance but no one knows how or why it is produced.

Certain things are quite definitely proved with regard to parathyreoprive and infantile tetany upon which most of the experimental work has been done. One outstanding fact is that the calcium content of the blood is decreased, and the other that if this be restored to some degree by introducing a soluble calcium salt, the symptoms of tetany disappear and the animal remains normal as long as the calcium level is kept high enough. These two facts were first clearly demonstrated by MacCallum and Voegtlin although some evidence as to the decreased calcium content of blood and tissues had been brought forward by European workers (Quest, Sabbatani and others). Exact studies proving the similarity in the reduction of calcium in the case of infantile tetany have been made by Howland and his assistants, Marriott, Kramer, and others. The truth of both statements has been confirmed by a host of other workers and there is now no doubt as to their correctness. It seems unnecessary to mention again the names of one or two investigators who have figured for years in the literature as having had results contrary to these.

As to the sodium and potassium content of the blood there are few definite statements but Howland and Kramer find by accurate methods which were devised for the purpose, that these substances are present in essentially normal amounts. Several authors have turned their attention especially to the sodium and potassium relative to the calcium and magesium on the basis of J. Loeb's studies of the importance of these relations in connection with the excitability of the nerve muscle preparation. This is indeed so well established an idea that it is brought forward as a final explanation of the electrical hyperexcitability whatever be the mechanism of the disturbance in the relations. However, the conditions are so complicated by the various anions concerned and by the varying rates of excretion of the alkaline kations that it is not always easy to refer the results of experiments directly to changes in the relations of sodium and potassium to calcium and magnesium.

Joseph and Meltzer thought that tetany could be cured by large injections of concentrated sodium chloride solutions and Voegtlin and MacCallum found that a slight improvement in symptoms could be produced in this way, lasting, however, only a short time, not accompanied by a lowering of the electrical excitability of the nerves and not in any way comparable with the immediate and complete removal of all the effects of tetany which follows the injection of calcium. Potassium acetate in their hands rather increased the intensity of the tetany and so did sodium bicarbonate. It is well known that injections of sodium bicarbonate in normal animals will greatly increase the electrical excitability of the nerves and produce convulsions not precisely comparable with the twitchings of tetany, and that this is accompanied by an increase in the alkaline reserve of the blood and the concentration of bicarbonate although all these changes shortly disappear.

It is not clear that an increased electrical excitability of the nerves or any symptoms resembling those of tetany can be directly brought about by attempts to increase the ratio of sodium and potassium to the calcium in the circulating blood although this disturbed ratio evidently exists when in tetany the sodium and potassium are in normal proportions and the calcium decreased. Possibly this is due to the rapid compensation by excretion of these substances.

With regard to phosphorus nearly all investigators agree that in the course of tetany there is or may be a retention in the blood. Greenwald found a distinct increase in the phosphorus content of the blood of dogs in tetany and Elias and Weiss, and Elias and Spiegel confirm this. Bluhdorn states that the phosphorus metabolism runs parallel with the calcium balance. Howland and his associates who find phosphorus markedly reduced in rickets, find it normal or increased in tetany and Freudenberg and György make the same statement.

This increase in phosphorus has led to a great deal of study of the effects of injecting phosphates into the circulating blood and in these experiments while there have been very contradictory results, it really seems that new light may have been shed on the whole chemical situation in the blood in tetany.

Binger found that the injection of phosphates in sufficient amount would cause a decrease in the calcium content of the blood and produce tetany provided alkaline or neutral phosphates were used. Mono-

sodium phosphate with its acid reaction caused a decrease in the calcium but no tetany and for this he offered no explanation. Jeppson also found that alkaliphosphates which are so abundant in cows' milk produce an electric hyperexcitability, especially in the case of the potassium salts. He is unwilling to believe that this depends upon changes in the calcium but thinks rather of a specific action of the HPO_4 ion together with that of the potassium but it seems that this conclusion is based upon some observations which are not in accord with those of other workers, such for example as the argument that salts of potassium are more effective than those of sodium because sodium bicarbonate does not affect the electrical excitability of the nerves. (On the contrary it has been repeatedly observed that the injection of sodium bicarbonate will produce a sort of tetany (Howland and Marriott, Harrop and others) and MacCallum has shown that upon the injection of sodium bicarbonate in normal dogs the electrical excitability is greatly increased although the exact symptoms of tetany are not reproduced.)

Tisdall injected dibasic sodium phosphate and found a reduction of calcium from 10.5 to 6.2 mgm. per cent, an increase of inorganic phosphorus from 5 to 19 mgm. and a change of pH from 7.4 to 7.5. There was no change in the sodium or potassium or chlorine concentration and none in the CO_2 combining power. After injection of phosphoric acid the changes are the same except that the reaction of the blood tends toward the acid side and there is reduction of sodium and the CO_2 combining power. He thinks the ratio of sodium to calcium is important and found one patient with tetany in which this ratio was diminished.

Elias and his associates found the phosphorus increased in the blood in tetany and observed that injections of primary as well as secondary phosphates increased the electric excitability with no change in the alkali reserve. He found that sodium bicarbonate decreased the symptoms of tetany while monosodium phosphate made them worse and concluded that there must be acidosis in tetany. In these conclusions he differs from almost all other authors.

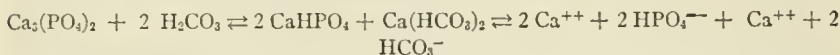
Porges and Adlersburg find that alkaline sodium phosphate produces tetany, while the acid phosphate does not, and a neutral mixture of alkaline and acid phosphates has no effect. Ammonium phosphate

($\text{NH}_4\text{H}_2\text{PO}_4$) removes the symptoms of tetany just as has been shown to be true for ammonium chloride. The effect of the phosphate is different according to the mode of administration for the acid action appears first and is transient, being succeeded two or three hours later by the phosphate action after the effect of the acid has been compensated. When given by mouth the acid effect is predominant and lasts longer.

Freudenberg and György in a large number of papers devoted mostly to theoretical considerations, state that in tetany there is an alkalotic tendency—that the essential element in tetany is the concentration of ionized calcium in the blood and that increased phosphates alone cannot produce tetany—acid phosphates do not tend to do so because they do not influence the dissociation of the calcium. They hold to the thesis that idiopathic tetany at least is due to the coincidence of an alkalotic metabolism and the retention of phosphates because under these circumstances the ionized calcium decreases when the phosphate increases and especially when the tendency is toward the alkaline side. Adlersburg and Porges, in their last paper, describe experiments on normal human adults in whom the intravenous injection of large amounts of NaH_2PO_4 produces an increase in electric excitability. This seems a very reasonable paper based on actual experiments in which they show that when acid sodium phosphate is injected into the veins in large quantity, an increase in the electric excitability appears at once even in the presence of an acidosis (cf. Elias and Kornfeld) and that this is heightened two or three hours later as the acid is compensated and the effect of the phosphate persists. They think that while the acid tends to decrease and the phosphate to increase electric excitability, one must keep in mind the quantitative relation of these substances, and that although it may be possible to explain the effect, as Freudenberg and György maintain, on the basis of the degree of ionisation of calcium, it is conceivable that anions as well as cations may have specific effects.

Howland, in his recent review of rickets, quotes from his previous paper concerning work of himself and Kramer with regard to the ionization of calcium. The solubility product constant explained with relation to silver chloride is equally applicable to calcium phosphate.

Tricalcium phosphate, $\text{Ca}_3(\text{PO}_4)_2$, is a substance soluble to only a slight degree. In the serum, tricalcium phosphate is subjected to the influence of carbonic acid which is present in considerable concentration. The formula may be written as follows:



If calcium phosphate is exposed to the action of carbonic acid an equilibrium is established between dicalcium phosphate and calcium bicarbonate and between these and calcium, HPO_4^{--} and HCO_3^- ions. When CO_2 tension of this solution is reduced a reversal of the process takes place and tricalcium phosphate is precipitated again. The reduction of the CO_2 tension in a solution containing HCO_3^- ions is equivalent to increasing the pH, i.e., the alkalinity.

Ross, working with Howland, refers, as do so many others, to the formula of Rona and Takahashi which shows that the calcium ion concentration depends not only upon the actual calcium concentration but also upon the bicarbonate ion concentration and upon the hydrogen ion concentration.

$$\frac{(\text{Ca}^{++}) (\text{HCO}_3^-)}{(\text{H}^+)} = K$$

Under these circumstances the concentration of the calcium ions would decrease as the bicarbonate ions increased but increase as the hydrogen ions increased and vice versa. A more useful conception is that employed by Freudenberg and György and others, although it is conceivable that even this is not yet complete.

$$\frac{(\text{Ca}^{++}) (\text{HCO}_3^-) (\text{HPO}_4)}{(\text{H}^+)} = K$$

From this the ideas so frequently expressed as to the importance of the ionised calcium can be readily compared with the actual results of experimental injection of bicarbonates and phosphates and changes in the hydrogen ion concentration although this not altogether theoretical "equation" does not bear directly upon the antagonism of other kations. It renders easy the explanation of the results of Adlersburg and Porges according to which a simultaneous increase in the phosphate ions and the hydrogen ions must result in changes in the calcium

ion according to their relative mass. An increase in the HPO_4 ion will decrease the available Ca ions unless at the same time the hydrogen ions are increased in equivalent degree, while naturally the injection of an alkali phosphate decreasing the hydrogen ion and increasing the HPO_4 ion will inevitably lead to a decrease in the calcium ions. It is evident that all of this admits as its underlying idea the essential importance of the calcium ion in its effect upon the electric excitability of the nerves, and that the various kations merely play upon it to decrease or increase it but this is in turn by no means incompatible with the still more fundamental possibility that it is the relative proportion of the sodium, potassium, calcium and magnesium ions which finally affect the excitability of the nerve—even though the change may be exclusively in the calcium.

It seems that the most important need at present in the study of the chemistry of tetany is some method whereby the ionized calcium may be actually measured in the blood. This has been so difficult that there are practically no observations upon this point. If we could directly measure the calcium ion content and the hydrogen ion content of the blood in normal animals and in those brought into tetany by any of the numerous ways known to be possible, we could establish objectively the accuracy of the equation just given and from that deduce and later confirm all the possible situations with regard to other forms of tetany.

It is at least clear already from this equation, if it is a correct representation of the actual conditions, that no change in the hydrogen ion concentration of the blood is necessary for the reduction of the calcium ions, since this can be effected by a corresponding increase in the phosphate ion or in the bicarbonate ion. On the other hand the equation would suggest that if a decrease in the calcium ion is found, we should expect at once an increase in either bicarbonate or phosphate ions. Whether this is true in all forms of tetany remains to be seen. It is clear that phosphorus is retained in parathyroid tetany and bicarbonate in gastric tetany and in that produced by the injection of bicarbonate. But in infantile tetany the calcium is decreased, the bicarbonate is not increased, the inorganic phosphorus is normal and there is nothing to suggest an alkalosis. It is probable therefore that other ions must enter into the equation here and indeed we do not know the mechanism of the definite decrease in calcium in infantile tetany in the presence of normal relations of the other ions.

In all these formulations of the chemical balance in the blood, the very important fact of the presence of numerous and complex protein substances is left out of consideration, although it is altogether probable that they may exercise a profound influence and perhaps control the situation to a great extent.

The study of Trendelenburg and Goebel upon the relative effect of normal and tetany blood upon the heart of a frog is generally referred to as a proof of the prime importance of the ionized calcium as such in affecting the electrical excitability of the nerves, but it does not seem clear that this is proved by their study. They found that normal blood when perfused through a frog's heart maintained a beat of a certain height and that blood from an animal in tetany allowed the heart to beat only half as powerfully although the normal beat was restored when normal blood was made to pass through again. Then they ashed both bloods and found that a solution of the ash of normal blood when perfused maintained a normal beat while that of the ash of tetany blood still allowed the heart to beat only half as strongly. If we assume that the ashing of the blood reduced all the calcium to the ionized state, the production of a normal beat by the ashed tetany blood would depend upon what proportion of the total calcium was in the ionized state in the normal blood. If the total calcium in tetany blood is only half that in the normal, it would seem that unless more than half of that in the normal was in the ionized state and in that condition necessary to the normal beat, there should be enough ionized calcium in the tetany blood ash to supply this requirement. It is evident that there are many possibilities and that it is by no means clear that this experiment proves the importance of the ionized state. It would be easier to assume from that alone that it is the reduction in the total amount of calcium in the blood which makes the tetany blood unable to maintain the normal beat.

Ross states very clearly the situation with regard to "three types of tetany, gastric tetany, tetany following bicarbonate injections and infantile tetany in which we meet with a common symptom although the blood of the patients shows a totally different picture.

In infantile tetany the calcium concentration of the serum is invariably reduced, in gastric tetany it is normal, whereas in tetany following bicarbonate injection it may be normal or reduced. In infantile tetany the

bicarbonate of the blood is normal, in gastric tetany it is greatly increased, in tetany following bicarbonate administration it is increased. In infantile tetany the chlorine content of the blood is normal, in gastric tetany it is greatly reduced and in the tetany following bicarbonate administration it may be reduced. In infantile tetany there is no evidence to suggest an alkalosis, in gastric tetany there may be a slight change in the pH of the blood toward the alkaline side. The phosphorus (inorganic) concentration of the serum is normal in infantile tetany. It may be increased in gastric tetany and also in the tetany following bicarbonate administration. The sodium concentration of the serum is normal in infantile tetany, in gastric tetany it is reduced."

These conditions may be shown in tabular form (see page 148) and to them there may be added other types of tetany.

From this table it is seen that with regard to some forms such as gastric tetany, bicarbonate and phosphate tetany and perhaps that following hyperpnea, our information is such as to suggest an explanation by way of the formula of Rona and Takahashi if we assume that the ionized calcium is reduced in the presence of an increase in either bicarbonate or phosphate ions. In infantile tetany and that following destruction of the parathyroids we have a great decrease in the total calcium of the blood but there is really nothing to explain it that is based on any firm foundation. It is true that there have been statements to the effect that there is an alkalosis in parathyreoprive tetany but these are contradicted by others on the evidence of equally good experimental work and no one has found an increased bicarbonate content in the blood. Inorganic phosphorus is retained in increased amount in experimental parathyroid tetany but not as a rule in infantile tetany and this suggests that the hydrogen ion concentration of the blood should be more carefully studied. At best, however, this would give only incidental evidence as to the condition of the calcium and none as to the actual reason for the alteration in its concentration in the blood. In attempting to explain infantile tetany in children fed on cows' milk, Orgler suggests that the excess of potassium and of alkaliphosphates is at work, and Wernstedt likewise regards the potassium the chief exciting cause. Human milk, according to him, contains more potassium in proportion to calcium but cow's milk brings much phosphate and probably lowers the calcium proportion.

Types of tetany

	CALCIUM	BICARBONATE OF BLOOD	CHLORINE	pH	PHOSPHORUS	SODIUM	POTASSIUM
Infantile tetany.....	Reduced	Normal	Normal	Normal	Normal or increased	Normal	Normal or slightly increased
Gastric tetany.....	Normal	Increased	Reduced	Normal to alkaline	Increased	Reduced?	Normal?
Bicarbonate tetany.....	Normal or reduced	Increased	Reduced?	Normal to alkaline	Increased		Normal?
Parathyroid tetany.....	Reduced	Normal		Normal	Increased		Increased
Incl. post-operative tetany.....	Reduced	Normal					
Hyperpnea tetany.....	Normal?	Increased		Normal to alkaline			
Phosphate tetany.....	Reduced	Normal	No change or increased	Normal to alkaline	Increased	Normal	Normal
Guanidin tetany.....	Reduced?				Increased		

Larsson isolated the proteins of whey and found them separately administered without any effect upon children in latent tetany but the milk plasma freed of protein stirred up symptoms of tetany.

Wetzel, on the contrary, could produce no increased excitability of the nerves with potassium salts and thinks that cows' milk does not produce ill effects on account of its potassium content. Gross and Underhill think that parathyroidectomy chiefly disturbs the relations of potassium and calcium with definite lowering of the calcium. Frank, Notham and Guttman also emphasise the importance of potassium in this regard.

Various pathological and physiological processes are found to accentuate tetany or to make manifest latent tetany. Howland and Marriott, and Ross have shown that in severe nephritis there may be a retention of phosphates and a lowering of calcium which would favor the appearance of tetany. Pregnancy, according to Stenvers and others, tends to ameliorate tetany and has no bad effect upon postoperative tetany but may influence it favorably. It is conceivable that this might in the late stages of pregnancy be partly due to the activity of the parathyroids of the child. This favorable influence is lost in lactation. This has long been known and it seems probable that it is to be explained as due to the removal of calcium from the body in the formation of the milk.

On the other hand, long continued tetany becomes associated with various pathological changes which seem more or less directly related to the calcium metabolism. Erdheim's studies of the dental changes and bone changes in rats deprived of their parathyroids are well known. Korenchevsky criticises these results by saying that diets deficient in calcium and vitamine A, or both, produce analogous changes and suggests that Erdheim paid no attention to the diet. Sainton-Poron describes chronic tetany with changes in the dental enamel, loss of hair, changes in the nails and cataract. Even the cataract may well be associated with disturbance in the calcium metabolism.

From still another point of view there have been a few observations on the relations of the parathyroids to disturbed calcium metabolism. Thus, Klemperer found enlarged parathyroids in a case of carcinoma of the breast with extensive bone metastases with osteoid tissue for-

mation. He connects the enlargement with a vain attempt on their part to keep pace with the disturbance in the calcium relations following destruction of the bone. Similarly, Günther found tumor-like enlargement of the parathyroids associated with extensive destruction of bone by the so-called multiple brown giant-cell sarcoma of bone which are really chronic inflammatory changes. Strauch found such tumor-like enlargements associated with osteomalacia. Such scattered observations are difficult to collate precisely but in each case it seems that there is an intimate relation of some sort with calcium metabolism.

Through much of the literature there runs the idea that there must be a toxin responsible for the tetany even though with the best will no one has ever been able to demonstrate the existence of any such toxin. The argument is invariably offered that since in experimental tetany the removal of a quantity of blood and its replacement by calcium free salt solution will stop the symptoms of tetany, there must be a poisonous substance in the blood which is now partly removed and for the rest diluted. It is true that the electric excitability of the nerves may be lowered and the symptoms disappear but it is not necessarily the effect of removing a toxin, for in a normal animal exactly the same lowering of the electric excitability occurs. The cure comes, no doubt, because the functional capacity of the nerves to produce a muscular twitching is decreased by their malnutrition and if it were allowed to go on without compensation it would become impossible in a very short time for any impulse to produce a twitching of the muscle. If in an isolated extremity the perfusion fluid instead of the purest blood is blood mixed with salt solution or Ringer's solution, the excitability of the nerves sinks to zero in a very short time. Thus, from that point of view there is little to show that a toxin is removed by bleeding the animal and replacing the blood with salt solution.

The reviewer has the impression that the arguments put forward in favor of the idea that the formation of methyl guanidin in the body is the actual cause of tetany are based largely upon the feeling of the need of finding such a toxic substance. Guanidin or methyl guanidin found in blood or urine and extracted by very complicated chemical procedures has an air of artificiality and at best the fact that the injection of such substances produces symptoms something like those

of tetany is not proof that it is the actual cause of tetany. Koch was the first to suggest this connection and was followed by D. Noel Paton, Findlay and others, who after making extended studies of the calcium deficiency of the blood and the mechanism of nervous hyperexcitability, accepted methyl guanidin as the substance produced from muscle by the decomposition of creatin which became responsible for the muscular twitchings. They found that guanidin and methyl guanidin when injected produced symptoms extremely similar to those of tetany, including the heightening of the electrical excitability of the nerves and that calcium acted to antagonize these symptoms as in tetany. Burns and Sharpe claim to have isolated from blood and urine of animals in tetany, and from the urine of children suffering from idiopathic tetany, guanidin and methyl guanidin in much larger quantities than are found in normal animals or children.

The other papers upon this bring various points of view. Henderson finds that after parathyroidectomy there is a fall in the total and free guanidin of muscle and a corresponding rise in creatin and thinks the excess of guanidin in blood and urine is due to its liberation from the muscle or failure of muscle to take it up from elsewhere. Burns and Watson thought that parathyroid tetany and guanidine poisoning, both of which cause interference with vago-cardiac inhibition, differ only in degree. In both calcium salts remove the effect upon the heart. Guanidine has a nicotine-like action poisoning synapses and in larger doses an atropine-like action causing paralysis of terminal ganglia in the heart of the frog. It does not paralyze the sympathetic accelerator mechanism. Natrass and Sharpe come to similar results. Bayer showed that guanidine injections produce a lowering of calcium in the blood approaching that of parathyroid tetany, but Klinger shows that in rats in guanidine poisoning the character of the symptoms is very different from parathyroid tetany and is not cured by calcium. There is bronchial spasm, difficult breathing, salivation, pruritus of nose and convulsions. In the cat guanidine produces vomiting, increase in motor and psychic excitability, restlessness, twitching and salivation, and it is here too most striking that while calcium salts cure parathyroid tetany they are useless in guanidine poisoning. Nelken found that guanidine may or may not lessen the calcium content of the blood but usually heightens the phosphorus content. Kummer, who

observed tetany produced by citrates and even by oranges and lemons, thinks that in tetany the loss of calcium leaves the muscle permeable to such poisons as guanidine. Frank, who studied chronic and latent tetany, assumed that it might be caused by an auto-intoxication with dimethylguanidine produced by splitting off from creatinin. Dragstedt and his associates are convinced that the poisons which produce tetany arise in the gastrointestinal tract through the activity of proteolytic intestinal bacteria and are protein split products (amines). The parathyroid prevents intoxication as it is part of a detoxicating system which includes the liver. Proper attention to the diet and the prevention of intestinal toxemia should prevent tetany.

The injection of guanidine, methyl guanidine, histamine, trimethylamine, etc., failed to produce typical tetany but there were vomiting, tremors, etc.—more easily produced in parathyroidectomized dogs than in normal ones. Luckhardt and Rosenbloom proceed on the idea that if guanidine is the poison in producing tetany, active diuresis should wash it out and cure tetany. Abundant injection of Ringer's solution even without calcium will cure tetany. There are many possible explanations of this without recourse to the necessity for a toxin—for example, phosphates may be washed away leaving the available calcium increased.

On the whole, the evidence in favor of the primary rôle of guanidine or methylguanidine in the production of tetany seems unconvincing though it would be so satisfying to find a single toxic cause of tetany.

Recent experience in the treatment of tetany is almost all of a nature to throw some light on the mechanism of its production and is therefore important in the study of the pathogenesis. Practically all authors agree that the administration of soluble salts of calcium by intravenous injection or in any way which insures the rapid absorption of the calcium will stop as though by magic all the symptoms of tetany. This was thoroughly demonstrated by MacCallum and his co-workers and the importance of calcium in connection with the hyperexcitability of the nerves shown by perfusing an isolated extremity with blood from which calcium had been in part removed by dialysis. Such nerves became abnormally excitable but were restored to the normal state when perfusion was continued with normal blood. Clinically, the immediate relief of symptoms upon intravenous injec-

tion of calcium salts has been everywhere confirmed, for example by Bluhdorn, Brown, MacLachlan and Simpson, Kneschke, Maggiore, Leicher, Beumer, Luckhardt and Goldberg, Howland and his associates, to mention only a few of those who have commented on it. Salvesen, in dogs kept alive for a long time with calcium, found that after the lapse of weeks there occurs an adaptation to a lower requirement of calcium and tetany remains in abeyance even though the content of calcium in the blood is low. Such dogs thrive on milk but develop tetany at once if given a meat diet instead of the milk. So, too, if calcium is removed from the milk by the addition of oxalates in proper amount they develop tetany at once, but if they are given a sufficient daily dose of calcium they are found not especially to require milk but thrive on a meat diet. The adaptation to the low level of calcium is not explained but the prime importance of sufficient calcium in the diet is perfectly demonstrated. Luckhardt and Goldberg reached quite the same result and found that at first the intake of calcium must be 1.5 grams per kilo daily. In their dogs upon a threshold diet of calcium, tetany recurred during the oestral cycle and no doubt would do so also during lactation.

In a more indirect way it is possible to cure tetany temporarily by the administration of substances which supposedly affect the ionization of calcium. Thus, as has been stated, Freudenberg and György recommend ammonium chloride as producing a tendency to acidosis. Porges and Adlersburg and Raab put their faith rather in ammonium phosphate. Bluhdorn approves of this but thinks of course calcium lactate and acetate more lasting in their effects than ammonium chloride or phosphate since the calcium is replaced. The bearing of these observations has already been explained. In gastric tetany MacCallum and his associates found that sodium chloride in large quantities prevented or cured the condition temporarily and McCann has since used with success ammonium chloride. Gamble and Ross found that by the use of sodium chloride the base is maintained in the blood and dogs lived longer than when treated with ammonium chloride.

Quite mysterious are the remarkable effects of exposure to the ultra-violet rays of the quartz lamp which has been shown by Sachs, Casparis and Kramer, Huldshinsky and others to cure the symptoms of

tetany and to raise the calcium concentration in the blood and also that of the organic phosphorus. It seems that this is due to increased absorption of these elements from the gastrointestinal tract (Orr, Holt, Wilkins and Boone). It is clear from this that rickets should be cured in the same way and it is well known that this is true. Little attention seems to have been devoted to the effect upon tetany of the other great cure for rickets, cod liver oil. Its action is also obscure and apparently due to an improved absorption and decreased excretion of calcium and phosphates. When rickets is cured the further symptom, tetany, is prevented or cured and in this sense cod liver oil is effective in tetany, but it is hardly possible to expect it to serve as an emergency treatment in cases of severe tetany.

Extracts of parathyroid glands have naturally been tried, both experimentally and clinically throughout many years and some of them seem to have some effect in restoring the calcium balance and the normal excitability of the nerves, but at best it is a slight and questionable effect and less satisfactory in experimental animals than in the tetany of adults, from which it may probably be assumed that the psychic effect of any treatment plays a part there. Doubtless it may be as it was before the discovery of insulin, that this is because the proper method of making an extract has not yet been found, but it seems that one might expect effects from large injections of a watery or salt solution extract of fresh parathyroids—yet as will be seen from curves showing the electrical excitability in parathyroidectomized animals (MacCallum and Vogel) the immediate effect is very slight and no cumulative action could be observed.

One supposes that it is for these reasons that clinicians confronted with severe tetany, whether idiopathic or post-operative, have recourse rather to calcium salts which act so promptly in the emergency, and may be used so satisfactorily to maintain health over a long period in chronic tetany. Nevertheless, surgeons have interested themselves recently to a great degree in efforts to implant parathyroid glands and this natural method of replacing one or more of those which have been destroyed has been successful in the hands of a few. It requires careful technique and the chance of success is at least greater when it is possible to make an auto-transplant—that is, to reimplant the patient's own parathyroids when they have been accidentally

removed. Dr. Halsted felt that this was the only chance of success but apparently others have succeeded with iso-transplantation. The other obvious requisite which is emphasized by Borchers is that the surgeon should recognize the parathyroid and not transplant lymph nodes or bits of thyroid. Roth describes a successful transplantation in a case of tetany during pregnancy and v. Eiselberg has had several cases in which he cured chronic tetany by this means.

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CERTAIN ASPECTS OF THE CHEMOTHERAPY OF PROTOZOAN AND BACTERIAL INFECTIONS¹

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The term chemotherapy in its more restricted sense is employed to designate that branch of investigation which deals with the discovery of chemical agents which act specifically on infectious diseases and with the study of their mode of action. The introduction of this term we owe to Ehrlich and the great progress which has been made in this field within the comparatively short space of two decades is due largely to the brilliant achievements and leadership of this worker. The subject is now so vast that in the short time which is allotted to us we cannot do more than discuss certain phases of what has been learned. Our treatment of the subject must naturally fall into two divisions, that dealing with the chemotherapy of bacterial infections and finally, that in which the animal parasites have been made the objective. We shall deal with the former first since efforts to influence experimental bacterial infections with chemical compounds antedate similar studies with animal parasites.

CHEMOTHERAPY OF BACTERIAL INFECTIONS

Not long after substances became known which would kill microorganisms in the test tube, attempts were made to apply such germicides to disinfection within the organism. All such experiments postulated a direct action of the chemical upon the microorganism within the host such as occurs in disinfection outside of the body. Among the very first investigations in this direction were those of Koch (1) who attempted to sterilize animals which had been infected with anthrax bacillus by the use of bichloride of mercury. Although he was able to inject this substance into the blood in such an amount

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as to achieve a concentration many times that which was necessary to kill the microorganisms *in vitro* all the attempts proved fruitless. Koch attributed the discrepancy between the *in vitro* and *in vivo* results to the fact that mercuric chloride forms a chemical union with the serum proteins as evidenced by the formation of a precipitate and was therefore rendered ineffective.

In later work, particularly that of von Behring, we see numerous other attempts to achieve internal disinfection. Occasionally indications were obtained with the use of certain inorganic substances but such results proved of no practical value because of their great uncertainty and irregularity and the great toxicity for the animal shown by the amounts of chemical required. Although occasional efforts in this direction continued, this type of investigation came to be regarded as futile and almost impossible of achievement. We can see in the words of von Behring an expression of this gloomy outlook. After ten years of persistent attempts he finally concludes:

It can be regarded almost as a law that the tissue cells of man and animal are many times more susceptible to the poisonous effects of disinfectants than any bacteria known at present. Therefore, before the antiseptic has a chance either to kill or to inhibit the growth of the bacteria in the blood or in the organs of the body, the infected animal itself will be killed. The pessimism of him who declared that disinfection in the living body is for all time impossible appears to be only too justified (2).

But with the more recent highly successful application of synthetic drugs to the treatment of protozoan diseases and because of the failure in so many instances of the more rational immunological methods, interest again centered in the possibility of the therapeutic application of bactericidal agents. The problem has proved extremely difficult to approach by any other means. It has been particularly difficult because of the lack of an empirical background such as had placed the qualities of arsenic, quinine and mercury at the disposal of the worker with protozoan infections. It has seemed to some that the only method short of the mere empirical testing of all sorts of substances in the experimental animal was to employ the bactericidal test at least as a means of initial orientation even though it appear the crudest makeshift.

The first extensive experiments in this direction were made by Bechhold and Ehrlich (3), who found in the group of halogenated phenols a number of substances which exhibited *in vitro* a high order of inhibiting action on the growth especially of the diphtheria bacillus. At the same time it was possible to follow certain influences which chemical constitution seemed to exert on antiseptic action such as the exalting effect up to certain limits of the multiplication of halogen groups or the addition of methyl groups to the molecule. But what is more important was the observation that none of these substances would sterilize laboratory animals infected with the diphtheria bacillus, and this, in spite of the fact that the toxicity of some of the substances was low enough to permit the achievement of a concentration in the blood 100 times that required to kill this bacillus in the test tube. A possible explanation for this failure was again found in the greater affinity of these disinfectants for the blood proteins than for the bacteria. Although no visible precipitate was caused on mixing their solutions with serum as in the case of mercuric chloride, their bactericidal action *in vitro* was greatly reduced by the process. In this work we find for the first time the expression "partial specificity" to designate a type of selective antiseptic action which a chemical may display towards a particular microörganism.

Compatibility with serum and partial specificity are factors upon which the subsequent workers with antiseptics have come to lay particular emphasis and their observations have furnished us with not a few examples of the latter. In the tendency toward specificity was found a grain of hope since it seemed a step in the right direction—in the direction of the specificity displayed by immunity principles. Many instances of partial specificity have been observed especially among the organic dyes. A number of these observations have been used as the basis of methods for the differentiation of bacteria in culture, such as that which employs gentian violet in media to restrain the growth of Gram-positive strains, or the use of brilliant green to separate the typhoid from the colon bacillus. A study of the bactericidal properties of the quaternary salts of hexamethylenetetramine (4) has brought out instances of substances which displayed a decided preference for the streptococcus. In not a few cases, substances were found which killed the streptococcus

in dilutions which were 20 to 30 times greater than those required to sterilize suspensions of meningococcus and gonococcus. A most unusual example of specificity has come to light in the case of the toxicity of the alkaloid optochin for the pneumococcus, but to this we shall return later. Quite recently Browning, Cohen and Gulbrausen (5) have uncovered perhaps a still more remarkable instance in the case of the cyanine dye, sensitol red. The lethal effect which it displays against staphylococcus is 2000 times that which it exhibits against the colon bacillus in peptone water. I am, however, not aware of any experiments which have been performed with this substance in infected animals.

In the quality of serum compatibility we have presented to us in its simplest form the idea that a substance must display a greater affinity for the microorganism than for the tissues of the infected animal in order to be germicidally efficacious. But few substances have been found which retain their full activity in the presence of serum. A few favorable instances have been observed among the quaternary salts of hexamethylenetetramine which were prepared from certain chloracetyl-amino compounds (4). A number of these exhibit a bactericidal effect in serum and protein solution which is very little less than that found in salt solutions, but their application to internal disinfection was without result. Optochin maintains toward the pneumococcus a high degree of bactericidal efficacy in serum. The low concentration of 1:20,000 in serum and even that of 1:8000 in oxalated blood have proved sufficient to kill this microorganism in two hours and in much higher dilutions it exhibits a definite growth restraining influence (6). However, this is considerably less than what is observed in ordinary salt solution since in the latter medium a dilution of 1:500,000 or more is all that is required. Much more favorable relationships have been observed in the case of the acridine compounds. Browning and his co-workers (7) have found that diaminoacridine and its methchloride, or proflavine and flavine respectively, actually exert a more potent action on the staphylococcus and colon bacillus in serum than in ordinary saline. This very unique quality and the unusually potent bactericidal properties of these substances led to their application in the local treatment of wound infections.

Still more difficult than to find substances which will resist the diverting influence of serum constituents is to find antiseptics which will maintain a high bactericidal effect in blood. Felton (8) made a number of observations in regard to this point, and but few instances of substances which retain a high order of antiseptic potency in this medium were observed. Mention may be made of malachite green and a few of its derivatives and especially the flavines and certain dyes of the rosinduline group. The observed values were, however, lower than those shown in ordinary saline.

There are still other properties which may be considered among the desiderata for the ideal internal disinfectant. One of these is speed of bactericidal action. It has been very difficult indeed to find substances which act rapidly under the conditions which obtain in the circulation. In order to kill the pneumococcus in fifteen minutes, optochin must be used in a concentration of 1:1000 in whole blood. Even flavine in the same concentration fails to sterilize in one hour although in two hours it may act in a dilution of 1:32,000. Taken alone speed of action may not be essential especially if a substance can be maintained sufficiently long in the circulation to develop slowly its action or to exercise merely a growth restraining or bacteriostatic effect. But here again we must postulate other conditions in order to obtain such a result. Repeated observations with optochin in animals and in man have shown that it is possible to render the blood antiseptic for pneumococcus but the observations of Moore and Chesney (9) have shown that this may be complicated by the development of fastness. Browning and Gulbransen (10) have demonstrated that the blood of rabbits may display a degree of inhibiting action for staphylococcus and the colon bacillus for several hours after the administration of diaminoacridine but this alone was apparently inadequate.

In the control of certain infectious processes the difficult requirements of proper distribution and tissue penetrability of the antiseptic must be taken into consideration. Lewis (11) in experimental studies on the chemotherapy of infections produced by the tubercle bacillus has stressed the importance of combining in a substance a degree of germicidal action with specific tissue penetrating power.

Finally, there is another group of requirements which must be

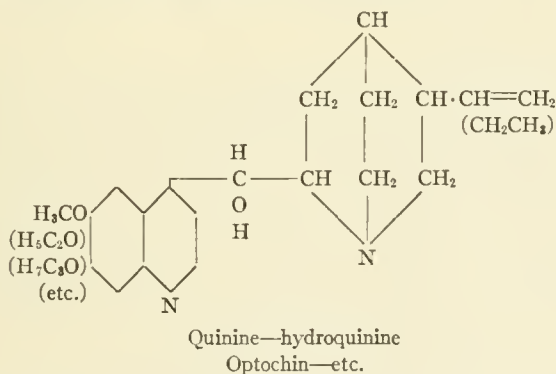
discussed in connection with the *in vivo* operation of bactericidal substances. Not only must a substance be of sufficiently low general toxicity but it must prove comparatively harmless to the general powers of resistance and to the normal protective mechanism of the infected animal. In this connection we may have to deal with quite an extensive and perhaps less tangible group of factors which can be incorporated into systematic study only with the greatest difficulty. It may be just at this point that the real problem lies and we shall have occasion to refer to this question later.

In this connection, observations have been made to determine the effect on phagocytosis which might be exhibited by bactericidal substances. The experiments of Felton (8) have recently shown that not a few highly bactericidal dyes and cinchona derivatives markedly reduce the phagocytic index in dilutions in which they might exhibit a bactericidal effect in blood. Exceptions in this regard may perhaps be the flavines which, according to Browning (7), are not appreciably injurious in dilutions above 1:500.

But if we consider in retrospect all of the conditions which may separate the observed *in vitro* properties of an antiseptic from its successful operation in an infected animal, it is apparent that we have presented a very difficult case for the idea that the control of bacterial infections may be approached from the standpoint of internal disinfection. This has certainly been very generally borne out in attempts to apply such substances in actual infections. It will be shown later that even in the more successful application of compounds to the control of protozoan infections, there have appeared repeated discrepancies between the action of substances on the parasite in the test tube and in the animal. It would seem, therefore, that for the time at least, we are thrown back on the empirical testing of substances in the infected animal as the only safe procedure.

The first striking instance of the successful application of a chemical to the treatment of a bacterial infection in the laboratory animal we owe to the observations of Morgenroth and his co-workers. Previous studies of Neufeld and others (12) had shown a certain biological similarity between trypanosomes, spirilla and the pneumococcus in

the susceptibility of their envelopes to the dissolving action of bile salts. While engaged in a study of the effect of quinine derivatives in experimental trypanosome infections in small animals, Morgenroth and Halberstädter (13) were able to demonstrate a certain curative effect of these substances. Then partly because of the possible biological similarity between the pneumococcus and the trypanosome and because of certain clinical observations on the applicability of quinine in the treatment of pneumonia, he was induced to make similar studies in experimental infections with the pneumococcus. Morgenroth and Levy (14) found that a pneumococcus septicemia induced in mice was but little affected by injections of quinine. On passing to hydroquinine which differs from it by but two hydrogen atoms a more definite influence was to be seen in delaying the death of the animals.



An optimal effect, however, greatly exceeding that of the former was furnished by ethylhydrocupreine, or optochin, in which the methoxyl group of hydroquinine has been changed to ethoxyl. It was found possible to save a good percentage of the infected animals by repeated injections, particularly of a solution of the drug in oil. Unfortunately, the clinical application of the drug has been limited by its untoward toxic properties, especially the tendency to produce amblyopia. Moore and Chesney (9), from extensive observations on patients suffering from acute lobar pneumonia, have concluded that the use of optochin in the treatment of this condition cannot be recommended since it is impossible to administer a sufficient

amount to produce an effective concentration in the blood stream without exposing the patient to the danger of toxic effects. During the use of the necessarily subeffective doses the pneumococci were observed even to become fast to the drug.

It would appear that Morgenroth in his first experiments was unaware of the potent germicidal action of this substance on the pneumococcus so that a knowledge of this property played no part in its initial chemotherapeutic application. We owe to Wright (15) the recognition of this quality as well as that of the fact that optochin kills the pneumococcus in high dilution in the presence of serum. The confirmation of these observations led Morgenroth (16) to attribute the curative action of optochin in the animal to its direct bactericidal effect and in his later studies he used the test tube as a guide. This later work developed the interesting influence which the chemical structure of the cinchona alkaloids exhibited on their specific antiseptic action.

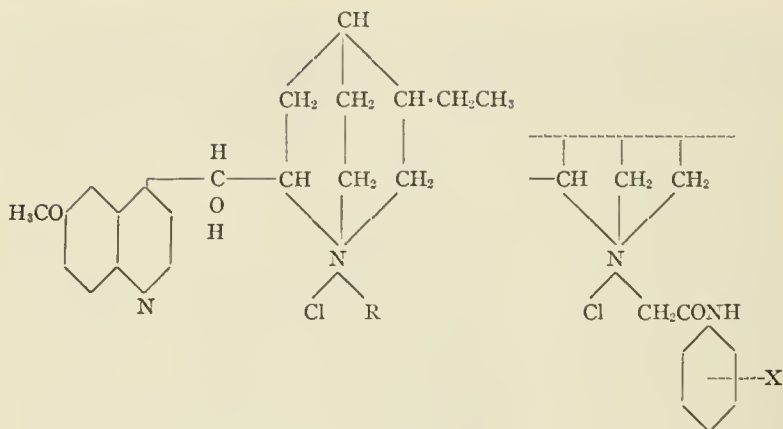
On passing from quinine, through hydroquinine to optochin, a sudden jump in the action upon the pneumococcus occurs only to fall again on passing higher up the series of alkyl compounds. However, with the stage of the isoamyl ether a bactericidal effectiveness towards streptococcus and staphylococcus becomes evident which is augmented on passing to the heptyl and isooctyl compounds and then drops again. These observations while interesting in themselves had to do rather with external disinfection and proved inapplicable to the treatment of systemic infections.

It would seem that the powerful bactericidal properties of optochin lends support to the belief that its curative effect in mice is a result of its direct bactericidal action. But certain observations of Neufeld and Engwer (17) and later of Moore (18) have suggested the interposition of the host in the process. Moore found that a single small dose of optochin in oil which in itself was quite ineffective in the treatment of an experimental pneumococcus septicemia in the mouse, when combined in the treatment with an amount of homologous antipneumococcus serum which was also far under the effective dose, was capable of increasing the threshold value of this serum at least fifty times. This result indicates that the combined action of the two therapeutic agents was many times that

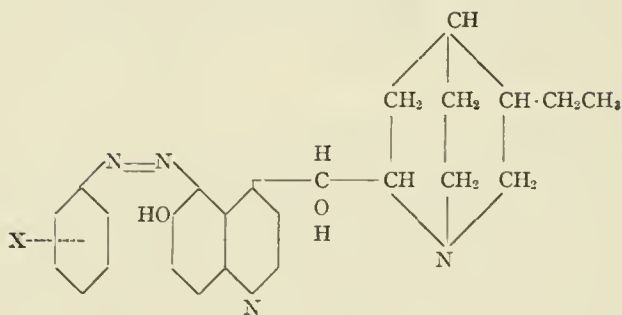
of a simple summation of their protective effects. Whether a similar synergism or rather fortification occurs between the drug and certain undetermined protective factors when the drug is used alone in a so-called curative dose has not been definitely ascertained but Moore's observations would seem to weaken somewhat the view that the bactericidal effect is the only factor.

Taking all that we have considered with reference to optochin and its near relatives as well as the ancient reputation which quinine has always had, we cannot escape the enticing suggestion that perhaps there is something inherent in the molecule of these alkaloids which, like the elements As and Hg, has conferred upon them certain potentialities as specific etiological remedies and that this property might be turned to wider usefulness if we could only find the way. A number of years ago Jacobs, Heidelberger, Wollstein and Felton turned their attention to this problem in the hope of finding new therapeutic agents for the treatment of infections produced by the pneumococcus.

The first problem was purely chemical. We have already seen what a pronounced change in biological properties has followed but slight chemical alterations in the quinine molecule. Would it be possible to find other means of altering the cinchona molecule in a systematic manner but in a way to maintain its general integrity and perhaps permit one to follow the influence of chemical modification on biological action? The naturally occurring members of the group as we have seen had been thoroughly studied and also their hydro derivatives and the homologous ethers of hydroquinine as well. A successful solution of the chemical problem was found by making use of the fact that these alkaloids as tertiary bases react with alkylhalides to form quaternary salts (19).



Quaternary salts of hydroquinine, etc.



Hydroxyazo dyes of hydrocupreine

In one direction in particular, by the use of chloroacetyl amino compounds as alkylhalides it was possible to develop if necessary a practically unlimited number of new derivatives and, in a way, to leave the original alkaloid molecule intact. Many substances were prepared into which a thorough selection of the usual organic groups had been introduced. Then again a similar opportunity was offered in the preparation of a series of hydroxyazo dyes from hydrocupreine (20), the phenolic compound obtained by demethylating hydroquinine. By coupling hydrocupreine with the diazonium salt of any aromatic amino compound it was possible to make a logical series of substances in which any chemical change could be featured at will. Still other series (21) of a similar character were prepared so that in a way it

might be said that the opportunity of testing the influence of chemical changes on the action of these alkaloids had been fairly completely presented. While realizing from all that was said before that the measure of bactericidal power was of doubtful importance, nevertheless, such studies were made because of their possible statistical value and since no other criterion short of the animal test was available. Within the groups of quaternary salts and azo dyes numerous instances of marked bactericidal power for the pneumococcus were noted and in several cases were found to equal even that of optochin. However, in the majority of instances, this activity was found to be greatly diminished in the presence of serum. Only the results obtained in the use of these substances in the experimental pneumococcus infection will be summarized here. Such tests made on substances, irrespective of bactericidal potency, showed a few which seemed to exhibit a small measure of therapeutic effect. The quaternary salts of hydroquinine with chloracetyl-*p*-amino phenol and chloroacetyl-*p*-anisidine in particular if given not too long after inoculation of the mice or if not in too large amounts often, though irregularly, cured the animals. An interesting and not infrequent observation which was made with these and similar substances was the effect of doses above a certain threshold even though much under the lethal dose. In such cases, instead of the apparent cure observed with small doses, an actual acceleration of the disease was noted. An explanation of this curious paradox is perhaps not difficult to find and may lie in the possibility that amounts of drug which are not frankly toxic may be sufficient to injure the animals natural powers of resistance in such a way as to make it more susceptible to the infection.

In this connection we have made also a survey of the various groups of organic dyes in regard to their curative effect on pneumococcus infections in laboratory animals but with similar unpromising results. Even flavine with its high bactericidal power in blood and serum for the pneumococcus gave but the slightest indications of influencing the infection. Observations made with quite a variety of other classes of substances were of a similar negative character.

Summary

While the experience of workers has brought to light not a few groups of bactericidal compounds, very few individual substances have displayed even a trace of specific therapeutic activity. These few substances have proved to be unique within the groups to which they belong since closely related compounds, or those differing from the active substances by relatively insignificant chemical modifications have proved to be quite useless. In regard to the general outlook for the chemotherapy of bacterial infections it may be said that at present the data available seem quite barren of suggestions. But I do not wish to imitate here quite the gloomy view which was quoted at the beginning of this paper. The mere instance of optochin and of the therapeutic value of chalmogroa oil and the more recently used esters of chalmogric and hydnocarpic acids obtained from this oil in the treatment of leprosy are indications that the control of bacterial infections by drugs is by no means impossible of achievement. The real difficulty lies in the necessarily opportunistic experimental method and the lack of a rational scientific means of approach. The problem is essentially a search for a substance of unknown nature but which must yet perform a service by a mechanism of which we are equally uncertain. It may be possible to inquire successfully into what biological properties such a substance must possess, or under what biological conditions it will have to operate; but such information will not point to the substance itself. In the case of bacterial infections, experience has presented us with too few points of contact between our knowledge of chemical structure and whatever biological properties may be required in the exhibition of a therapeutic effect to present a rational basis for the search for curative substances. Perhaps the real solution of the problem may be ultimately found by gaining an insight into the chemical nature and mode of operation of the substances which are normally elaborated by the organism in its fight against infection.

CHEMOTHERAPY OF PROTOZOAN INFECTIONS

In the application of chemical agents to the treatment of protozoan diseases we are confronted by a far more encouraging state of affairs.

We have already alluded to the traditional virtue of quinine and mercury and that these were purely empirical acquisitions. However, with the recognition of the etiological factors of the diseases which these substances influenced, the rôle which they occupied as real curative agents was likewise recognized. During the past twenty years considerably more has been added to our chemical equipment in the treatment of this group of diseases as a result of the application of laboratory methods of both chemistry and biology. In the present paper I can, however, touch upon only certain phases of this development. The appearance in 1902 of the classical experiments of Laveran and Mesnil (22) was the beginning. By the use of sodium arsenite these workers succeeded in temporarily clearing the blood of mice infected with the parasite responsible for the disease of cattle called "Nagana." Then there followed within a few years a succession of observations in which substances belonging to a number of different groups were found to possess a similar efficacy in experimental infections produced by the same or similar animal parasites. These substances fall naturally into several groups which it will serve our purpose best to treat separately. Soon after the observations of Laveran and Mesnil, Ehrlich and Shiga (23) demonstrated that mice infected with *Trypanosoma equinum* could be permanently cured by a single injection of trypan red, a tetrazo dye prepared by coupling tetrazotized benzidinesulfonic acid with a naphthylaminedisulfonic acid. This is noteworthy as the first instance of the cure of an experimental trypanosome infection with a single dose of a synthetic chemical. However, the value of the substance itself seems to have been limited, since it failed to exert a similar therapeutic effect in other animals or with other types of trypanosomes.

We meet here for the first time a peculiar paradox which was destined to appear repeatedly in later work. Although trypan red proved to be active *in vivo*, it was inactive in the test tube. At the time, these workers noted the staining of the tissues of the animal by the dye after injection. They considered that its action was possibly due to the persistent effect of small amounts of the drug or of some active alteration product of it upon the parasites. The production of an active immunity in the animal by the rapid destruction of the parasites by the drug was considered also as a contributing factor.

Observations of a similar character with other dyes became the basis for much subsequent discussion and inquiry as to the mechanism involved in the successful removal of trypanosome infections by such substances. In these discussions, which I cannot present at length, attempts were made to explain this paradoxical action also by the assumption of a direct injury produced by the dye on the trypanosomes of a character which did not affect their motility but prevented their multiplication. The suggestion that a chemical alteration of these substances in the body to a more active form, while not so obviously applicable to this class of substances, has been more successfully applied, as we shall see, in explaining the action of arsenic compounds. Subsequently, Nicolle and Mesnil (24) found from the study of a large number of similar substances a benzidine dye, trypan blue, which proved still more efficacious in that it cured mice infected with the more resistant trypanosomes of Nagana and Surra. Dyes of all kinds were then investigated and, in succession, members of the triphenylmethane group and a number of orthoquinoid dyes such as the pyronines, oxazines, thiazines and acridines were successfully brought into the field of investigation. But the practical application of the dyes, it would seem, has been on the whole rather restricted. The discovery of the therapeutic properties of the individual members of the different groups appears to have been for the most part the outcome of a rather opportunistic selection of substances. There have been attempts to correlate the therapeutic properties of some of them to their chemical constitution and to their selective staining property for the parasite. But on the surface at least, it would seem that but few facts have emerged which could be applied to a systematic attempt at the further chemical development of the dyes, or in explanation of whatever degree of therapeutic value they may have shown.

An exception to this, perhaps, is the rather striking property which was found by Ehrlich and his associates to accompany the presence of the orthoquinoid structure of certain dyes. These dyes quickly produced tolerant or fast strains of trypanosomes which were also quite fast to arsenic. Paraquinoid dyes did not show this effect. Again Nicolle and Mesnil (24) in a thorough survey of the tetrazo-benzidine dye group concluded that, in order to develop therapeutic

qualities, the coupler used must be a naphthalene derivative containing at least one amino group and two sulfonic acid groups. It will be seen in chart 1 that this condition is satisfied by both trypan red and trypan blue.

And it is possible that this may have been made the starting point for the preparation of a substance known as Bayer-205, which has recently come to offer considerable promise. The exact nature of this substance has been carefully withheld but there is a suspicion that it

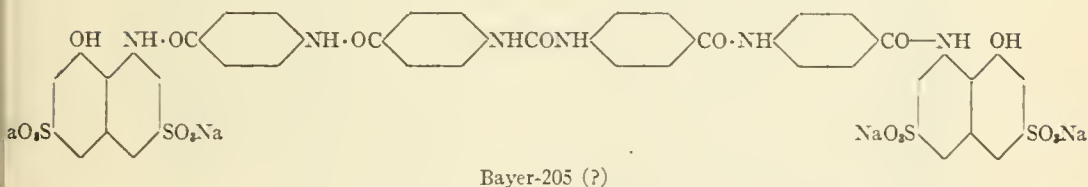
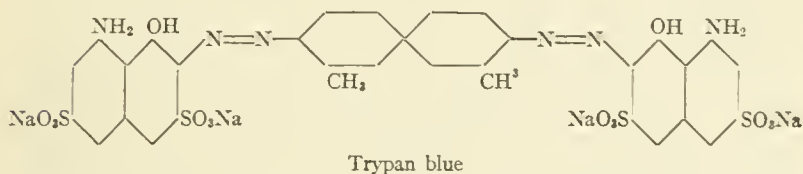
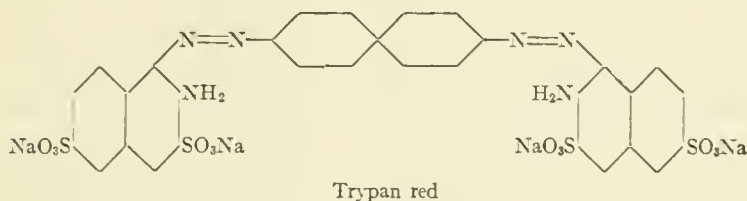


CHART 1. POSSIBLE RELATIONSHIP OF BAYER-205 TO TRYPAN RED AND TRYPAN BLUE

belongs to a group of compounds which the Bayer Company patented some time ago. The formulae which suggest the possible relationship of this substance to trypan red and trypan blue, (25) are shown in chart 1.*

Preliminary observations of Handel and Joetten (26) and Mayer and Zeiss (27) have shown that this substance possesses remarkable curative properties in experimental trypanosomiasis. In small doses which were often far below the toxic dose for the particular animal,

* A possible formula for this substance has been recently discussed by E. Fourneau and others. *Ann. d. l'Inst. Pasteur*, 38, 81 (1924).

the drug was found to cure the ordinary laboratory animals which had been infected with different strains of trypanosomes. Here again we find the curious circumstance that the substance itself displayed no marked action on the trypanosomes *in vitro* and microscopic studies seemed to suggest that the drug may act by preventing multiplication of the parasites. Later, therapeutic experiments on dourine in horses pointed to its usefulness as a prophylactic and curative agent but more important was its application to human trypanosomiasis. We shall return to a discussion of this in another connection. Although we can only infer the probable nature of this remedy, it would seem that an advance has been made by applying chemical knowledge to the problem of varying a group of substances which had already given indications of therapeutic value. Thus, the dyes from which this substance has emerged have come to share again, for the time at least, a prominence which was taken from them very early in chemotherapeutic studies by the advance in the application of arsenic compounds.

If we turn now to the arsenic group we find that the organic arsenicals have proved a most profitable direction for the experimental studies which led to the finding of synthetic specific remedies. This is attributable in part to the fact that we are dealing with substances in which the therapeutic quality is apparently inherent in the arsenic itself irrespective of the actual form or by what mechanism its action may come into play. This has made chemical reasoning simpler in attempting to refer biological action to chemical constitution, for it is far more difficult to interpret what should be the point of reference when a large organic molecule is considered with regard to any inherent biological activity.

The impetus to investigation in this direction came with the report of Thomas (28) that a proprietary arsenic preparation called atoxyl displayed a definite therapeutic effect in animals infected with different strains of trypanosomes. The rapid confirmation of this observation soon led to its employment in African sleeping sickness and it has proved helpful in the treatment of the early stages of this disease.

But a step of the most far-reaching importance was Ehrlich's (29) brilliant recognition of the true nature of atoxyl as the sodium salt of *p*-aminophenylarsonic acid or arsanilic acid. In analogy with

sulfanilic acid, it is formed by the fusion of aniline with arsenic acid. In its essentials, the chemistry of organic arsenic compounds had been pretty thoroughly studied by Michaelis and his co-workers but the methods at the disposal of these chemists were, in a sense, limited. The discovery that arsenic could be introduced into the molecule by direct arsenation opened to Ehrlich and his associates a means for a most thorough development of the chemistry of this group of substances. The immediate suggestion which the structure of *p*-aminophenylarsonic acid presented was detoxification by acetylation of the amino group. But the resulting product arsacetin, though possessed of a degree of therapeutic power, was soon dropped because in its clinical application, like atoxyl, it not infrequently produced blindness and other toxic manifestations. Here again the peculiar paradox appears that the pentavalent compounds are without appreciable action on the trypanosomes *in vitro*. But Ehrlich's (30) fertile imagination soon found a possible solution for this discrepancy. The fact was noted that individual animals treated with certain pentavalent compounds would show a varying tolerance for the drug and those which exhibited the lower tolerance could be cured by smaller doses. This observation was coupled with the well known ability of the tissues to exhibit a reducing action and with the fact that certain trivalent forms of arsenic are far more toxic than the pentavalent forms. Ehrlich concluded that the pentavalent arsenicals are reduced by the tissues and unfold their activity in the trivalent form. This view was supported by the direct comparison of the relative toxicities of the 3 stages of oxidation of arsanilic acid, *p*-amino-phenylarsin oxide, is 75 times more toxic for the mouse than arsanilic acid, while the arseno compound is 30 times more toxic. Similar toxicity relationships have been repeatedly observed.

But more important still is the contrast in the trypanocidal effect of the oxide and the arsonic acid *in vitro*. In a dilution of 1:100,000 the former kills the microorganisms immediately, whereas atoxyl fails to act even in a 5 per cent solution. In some cases the arseno-compounds have shown a similar potent action in the test tube but this has not always appeared. Mention may be made of observations of the failure of salvarsan to act directly upon the relapsing fever organism (31) or on the *Treponema pallidum* in the test tube although

the organisms thus treated are observed to lose appreciably in virulence as shown by subsequent injection into animals.

Ehrlich and Roehl (32) placed particular emphasis on the oxide stage which may be formed either by reduction of the pentavalent compound or by oxidation of the arseno derivative, as the form in which the biological manifestations eventually appear. It is perhaps important to mention this point since recent statements have appeared which seem to overlook this fact.

The conception of the biological potency of trivalent arsenic was at once applied to further synthetic attempts to find new curative agents in the arsenic group. The great toxicity of the arsenoxides however and the possibilities which the arseno compounds came to afford was the reason for the main interest which seemed to attach to the latter group. This work culminated in the brilliant discovery of the value of salvarsan and its derivatives in the treatment of certain spirochaetal diseases.

In his theoretical considerations to explain the action of chemotherapeutic agents, Ehrlich assumed a direct action of the substance, or its alteration product, on the parasite. He conceived of this action as being brought about by the avidity of certain groups or chemoreceptors in the cell of the parasite for certain groups contained in the chemical. By assuming these discrete affinities within the cell the attempt was made to give more visible expression to certain phenomena which were observed in the reaction of microorganisms to chemicals containing certain groups in the molecule. But on the whole workers no longer agree that such precise definition can be given to the relationship between chemical constitution and therapeutic action.

The efficacy of salvarsan was attributed in part to its orthamino-phenol group which was supposed to direct the drug at the parasite in a way to permit the lethal action of the arsenic to develop. The analogy was drawn with trypan blue which likewise possesses an amino and hydroxyl group in positions *ortho* to one another. But there are many cases where such analogies do not hold. It is said, for instance, that the isomers of salvarsan containing this configuration are all much less active than salvarsan itself. Again particular stress has been laid on the dystherapeutic effect of the methyl group but

numerous instances have arisen in which substances containing the methyl group have actually exceeded in effectiveness the unmethylated compound. In this respect trypanflavine is more efficacious as a trypanocide than the unmethylated proflavine. Quinine which is methyloxycinchonidine is much more efficient in malaria than cinchonidine or the phenol, cupreine.

The sulfuric group is still another well known example. In many combinations it may destroy therapeutic properties. Morphine or quinine when converted into the sulfuric esters although again easily recoverable by saponification, are physiologically practically inert. On the other hand several sulfuric acid radicals are contained in the molecule of trypan red, trypan blue and probably Bayer-205. The conclusion is forced therefore that we may attribute only the bare possibility of certain effects to certain groups in the molecule and in any event, the molecule as a whole certainly must be considered.

But the striking changes in biological properties that often follow very minute changes in structure and the occasional appearance of regularity of biological response to such changes have always been a fascination and a source of temptation to the worker to try to utilize such general tendencies in the synthesis of drugs.

Several years ago, a group, Brown and Pearce, in collaboration with Jacobs and Heidelberger, at the Rockefeller Institute, investigated a phase of this question. In this work the attempt was made to obtain some information from a systematic biological study of the toxic and therapeutic properties of certain types of arsenic compounds which might be applied to the finding of new remedies. This work which at first embraced a study of the treatment of experimental trypanosomiasis was later extended to include that of the experimental infections produced by the relapsing fever organism and of experimental syphilis in the rabbit.

We have already described the emphasis which has been placed on trivalent arsenicals. Since the biological activity was an exhibition of a property of some form of trivalent arsenic, it was undoubtedly a logical step if considered alone, to use this group of compounds directly. But we may regard this question from another angle. There seems fair evidence at hand that the greater tolerance for pentavalent arsenicals as a group is not alone a function of their greater biological

inertness as compared with the trivalent compounds but also to the speed with which the greater portion is excreted before the reducing action of the tissues can manifest itself. This is but a manifestation of the fact that the arsonic acids which form neutral salts possess a relatively greater portability in the blood stream and body fluids than the trivalent forms, as well as a greater penetrability and may readily and quickly diffuse through the tissues. The arsenoxides possess this to a much less degree and because of their degree of unsaturation are chemically very reactive and while perhaps initially portable, readily become bound by and denaturize such functional or tissue elements as may be determined by the general character of their molecule before the same degree of distribution can be achieved. This rapid fixation finds expression in the rapid exhibition of potent toxic manifestations. The arseno compounds as a group possess double the molecular weight and although perhaps also quite reactive, are the least readily transported and the least diffusible of all. When there is no special acid salt forming group, substances of this class may be held in solution for a short time only, aided perhaps by the protective serum colloids, but may be readily deposited as such along the route before complete distribution is possible and then more slowly unfold their toxic effects through oxidation to the oxides. If we were dealing with an infection which is confined to the blood stream alone it would seem therefore admissible to place reliance upon the direct quick action of the trivalent compounds and the simple formula of the ratio of parasitotropic to organotropic properties of a substance. But when the more usual tissue infection is to be influenced the question of distribution and diffusibility becomes a very important factor to consider. It is quite conceivable that certain advantages in this regard may result from the use of the pentavalent form which may compensate for what it may seem to lose in immediate direct parasitocidal properties. By more complete distribution and tissue penetration it might be possible to reach localities which may harbor such residues of an infection as may again become generally active. In such localities the reducing action of the tissues may supply a small but sufficient amount of therapeutically active material.

In spite of Ehrlich's somewhat orthodox adherence to the trivalent idea we can see evidence of his occasional return to the pentavalent

compounds. In his *Schlussbetrachtungen* (31), the possibility is pointed out of the applicability of hydroxy aminophenylarsonic acid, the pentavalent stage of salvarsan, for local therapy where the arseno compound circulating in the blood cannot reach the affected tissues in sufficient amount, as, for instance, in local eye involvement. Hata's (31) experiments showed several pentavalent compounds to exhibit a good ratio of curative to toxic action but we obtain the impression that these workers were perhaps prejudiced against the group because of a neurotropic tendency which a number of these substances displayed. This exhibited itself in the development of incoordination in the so-called "dancing-mice."

Considerations of a practical character have arisen in connection with the arseno compounds caused by certain inherent difficulties which are associated with their chemical and physical properties. They are unsaturated, highly reactive substances. Slight modifications in the method or conditions of preparation may have a great influence on their general toxic and also therapeutic behavior. It is not possible to obviate this readily by the subsequent use of the ordinary methods of purification such as recrystallization but very careful control of preparative conditions is required which can be only empirically ascertained. It has been necessary, therefore, to maintain a constant biological control of those substances of the group which are clinically employed. Further, the development of this class of substances is in a sense limited because the arseno compounds depend for solubility on the presence of salt forming groups. The pentavalent compounds possess much more favorable chemical and physical properties. The arsonic acids as a class are perfectly stable, crystalline compounds which can be readily purified. They form stable salts and therefore do not require the presence of other salt forming groups for solubility.

It appeared to us, therefore, that the development of pentavalent compounds was certainly worthy of further trial. In the selection of the chemical material for our studies such groups of substances were chosen which would combine a sufficient degree of accessibility with the opportunity, for ample and logical chemical development in such direction as the biological results might indicate. From this material it was hoped to accumulate statistical data regarding the influence of

chemical constitution on biological action which could be utilized in further work. An idea of the chemistry (33) of these groups of substances is best obtained by reference to chart 2. The studies were not alone confined to the pentavalent group but, in certain instances where the presence of a solubilizing group would permit, reduction to the trivalent stage was tried to test the value of such a chemical transformation.

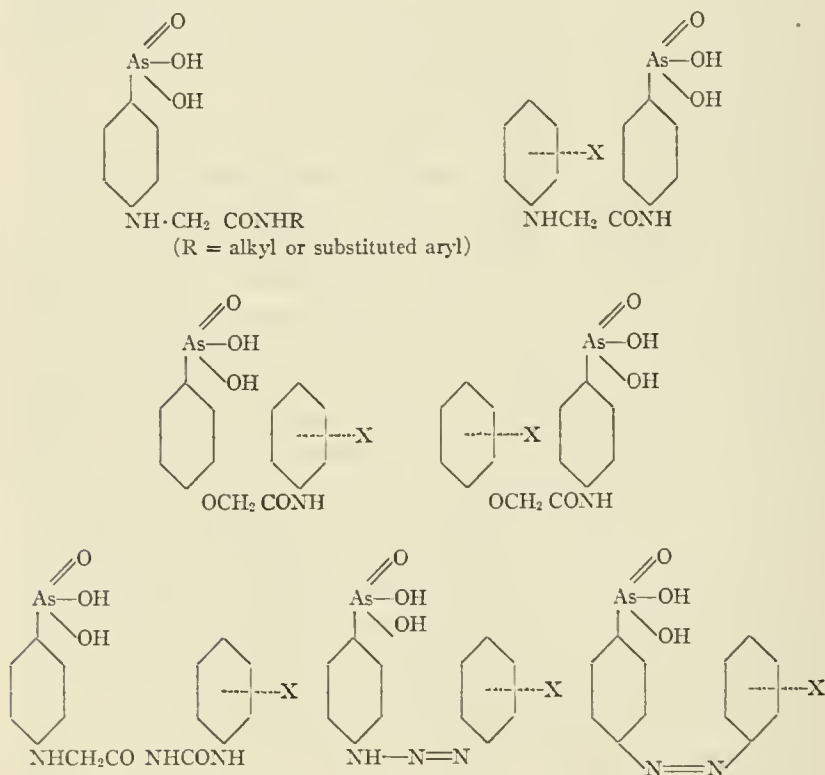


CHART 2

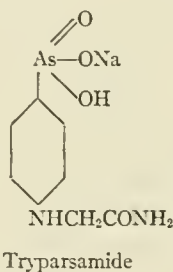
Over 230 substances were prepared. Their action as regards toxicity and therapeutic properties upon one or more laboratory animals infected with the different animal parasites was studied. The observations were too extensive to attempt to do more here than very briefly and superficially summarize the results. Substances were

obtained which varied all the way from those possessing a very high toxicity to those which were easily tolerated and at the same time exhibited varying degrees of therapeutic power. But the observations with these substances brought out the fact that in the biological measure of each compound so many different factors had to be considered that it became extremely difficult to find any sharp regularities with regard to the influence of structure on action. But in some cases, it was possible to note a few very general tendencies which were utilized to advantage in the work.

The most important group and that which was most fully studied was that of the amide and substituted amides of phenylglycine-*p*-arsonic acid (33). This type of compound was chosen partly for the reason that the glycineamide, $\text{NHCH}_2\text{CONH}_2$, group not only offered an excellent chance to add practically any chemical group to the molecule which was desired, but because the glycineamide group itself is a normal constituent of proteins and should be inherently compatible. These substances were in general prepared by the interaction of the amino group in arsanilic acid or analogues with chloroacetyl derivatives of amines in which members of both the aliphatic and aromatic series were given ample representation. Changes in both therapeutic and toxic effects were observed in response to chemical alterations in the molecule often of a decided character which depended not only upon the animal but also the type of infecting organism. Only in certain instances was it possible to follow any general tendencies. To begin with the parent substances, phenylglycine-*p*-arsonic acid, we have met a near relative before in the form of arsenophenylglycine, which is obtained from the arsonic acid by reduction. Contrary, however, to the potent therapeutic effects exhibited by arsenophenylglycine in experimental trypanosomiasis, the pentavalent form while it can be tolerated by laboratory animals in large amounts is practically devoid of therapeutic effect. In this series as far as it was carried the carboxyl group, while acting fairly constantly as a detoxifying influence, was found to possess a dystherapeutic effect. This was at variance with instances noted in other groups of compounds and like all such irregularities, which have made prediction impossible, there is no explanation at hand. In general, it has become evident that the tendencies which are observed in the

apparent influence of constitution on biological action are confined pretty narrowly to substances which are built on the same general plan.

On passing to the amide or ureides of the compounds containing these carboxyl groups, no matter where they were situated in the molecule, there was in general an appearance of therapeutic effect in spite of a tendency to increased toxicity. This therapeutic effect of course varied greatly with the particular compound, the animal and the infection used. A number of substances of the series from the biological standpoint proved to be of outstanding interest. In an extensive series of biological tests, Brown and Pearce (34) definitely established that one of these in particular possesses unusually favorable biological properties. This substance, the sodium salt of phenylglycineamide-*p*-arsonic acid, or tryparsamide as it has since been named, has now come to be of definite practical therapeutic importance.



Tryparsamide is a white, crystalline, stable substance which is extremely soluble in water with the formation of a practically neutral solution and is very easily prepared by the interaction of sodium arsanilate and chloroacetamide. The biological properties of this substance, are summarized by Pearce (35) as follows:

Toxicological experiments demonstrated that the reaction of different species of laboratory animals to the drug was of a favorable character. The substance lends itself well to almost any method of administration and can be given to animals in large doses and the toxic effects were confined to doses relatively close to the minimum lethal dose. The recovery of animals from sublethal intoxication was remarkably rapid and complete, thus making possible the repetition of large doses at comparatively short intervals of

time. The therapeutic activity of the drug in experimental trypanosomiasis was particularly evidenced by the relative speed and sharpness of action in the acute blood infections of mice and rats and by the potency and duration of action in the subacute and chronic tissue infections of guinea pigs and rabbits. The accomplishment of a permanent cure was obtained in the experimental infections produced by 5 strains of pathogenic trypanosomes: *Tr. brucei*, *Tr. gambiense*, *Tr. evansi*, *Tr. equiperdum* and *Tr. equinum*. Comparative experiments in laboratory animals with drugs which had been previously used such as atoxyl, arsacetin, arsenophenylglycine, and the salvarsan derivatives all of which had been previously used in the treatment of human trypanosomiasis showed that tryparsamide was in many respects superior.

In her clinical observations (35) made in the Belgian Congo several years ago, Pearce was able to substantiate the favorable indications which the experimental results with tryparsamide had given. It was shown that the drug is well tolerated and gives rise to no untoward symptoms except an occasional tendency, following too intensive a use of the drug, to produce visual disturbance in certain advanced cases. This, however, was usually transitory. The therapeutic results obtained in the preliminary use of the drug in 77 cases of sleeping sickness produced by *Trypanosoma gambiense* were very encouraging. More recently Chesterman (36) in the Belgian Congo has definitely confirmed Pearce's general observations. From observations on the intravenous use of the drug he concludes that the maximum tolerated dose (which he believes should not exceed 4 grams per week for the full-sized adult) if given regularly for a period of about eight weeks is capable of completely removing trypanosomes from and rendering normal, the cell content of the cerebrospinal fluid of even the most advanced cases. This change is accompanied by a very marked clinical improvement which was observed to persist for practically a year which was the longest time which had elapsed since his last use of the drug. Improvement was hardly less marked in cases which had resisted previous treatment with the drugs such as atoxyl. By a careful check on the patient it was possible to avoid the danger of any appreciable degree of visual disturbance. Up to the present, we see no reason to change our estimates of the value of the drug.

Smillie (37) has recently had the opportunity to extend the studies with Tryparsamide in another direction, to the treatment of mal de caderas which has become one of the biggest economic problems of the vast Paraguay Valley in South America, since this disease is causing the yearly loss of thousands of horses. In a preliminary communication he reports that the drug has been found to be definitely efficacious in the treatment of the early stages of the disease when the blood of the horse or mule is swarming with trypanosomes. This stage is from all practical considerations of the greatest importance to control.

But to return to human trypanosomiasis, it is seen that the two main directions which chemical efforts have taken to produce remedies for this disease have almost simultaneously yielded results which now give a more encouraging outlook with regard to its control,—on the one hand Bayer-205, most probably closely related to the benzidine dye group, and tryparsamide, a pentavalent arsenic compound. At the present time, it is perhaps too early to compare the relative efficacy of Bayer-205 and tryparsamide. There have been reports already with regard to Bayer-205 which indicate that the drug, although of definite value as a prophylactic or in the early stages of the disease, has failed to give results in the more advanced cases. Tryparsamide, on the other hand, as Pearce and Chesterman have shown, has been the cause of definite improvement in patients even in the more advanced stages of the disease. Bayer-205 has been reported to exhibit a definite toxic behavior from which the patients or animals only slowly recover. This takes the form occasionally of an albuminuria. A general tonic effect has characterized the behavior of tryparsamide and perhaps the only untoward action has been the occasional visual disturbance which has been noted to follow too intensive a use of the drug. But, as before stated, this is in most cases of a temporary character. We shall perhaps have to wait a number of years before these drugs will have reached their final evaluation.

There is a temptation perhaps to speculate with regard to the chance that the more successful of the arsenical remedies which have been used for the treatment of human trypanosomiasis have been pentavalent compounds. In the case of tryparsamide, there seems every reason to attribute its efficacy not only to its powerful stimulating

action on the animal economy and on the animal resistance but to its great tissue penetrability and to its affinity for the tissues of the central nervous system which are involved in the more advanced stages of the human disease. This property has been demonstrated again in the behavior of the drug in certain types of neurosyphilis. In neurosyphilis there is a distribution of organisms in the central nervous system which is in a sense comparable to that which occurs in trypanosomiasis. This has in the past suggested the possibility that perhaps even at the time when the etiological relationship of paresis to syphilis was merely suspected, drugs which had proven efficacious in the treatment of human trypanosomiasis might find a usefulness in the treatment of paresis. Years ago atoxyl, for instance, was repeatedly tried in this connection but in all these attempts it proved quite inadequate. It was, however, a natural thought to consider tryparsamide in this connection although its treponemacidal properties are not very marked. Nevertheless, in experiments in rabbits it had been shown to possess a marked action on the experimental lesions produced by the spirochetes. This was evidenced by their complete resolution and healing even in the presence of actively motile spirochetes which seemed to show little or no tendency to cause a recurrence.

Lorenz, Loevenhart, Bleckwenn and Hodges (38), in order to test the applicability of tryparsamide in this connection undertook a series of observations on a group of patients afflicted with paresis. They have concluded that tryparsamide injected intravenously is definitely efficacious in the treatment of early paresis and certain other forms of neurosyphilis. Later observations have essentially confirmed this. It is perhaps too early to make a more precise statement with regard to the clinical use in all of its phases of tryparsamide in this disease but sufficient is already known to demonstrate that it should have a field of usefulness, in certain conditions for which there has been heretofore no efficacious mode of treatment.

Only certain aspects of chemotherapy have been considered here but there are other phases of the progress which has been made in the application of chemical agents to the specific treatment of infectious diseases which might be reviewed with profit. But enough has been brought out to show that the results achieved have been far in excess

of what was needed to justify the initiation of this type of investigation. In no sense, however, can any result such as the arsphenamines, Bayer-205 or tryparsamide be considered the summit of possible attainment and there is every reason to expect the ultimate discovery of more powerful agents. And as time goes on, new groups of substances and an ever widening field for their therapeutic application may well be expected. There is perhaps no more hopeful direction for the continuation of effort. But those who engage in such undertaking must carry with them a certain measure of opportunism, since in spite of the constantly increasing number of positive observations, there is still lacking a general theory as to the chemical and physical factors which underly specific therapeutic action. In this respect we are perhaps less informed than in the case of certain rules which seem to govern pharmacodynamic action. But it is, perhaps, in this direction that further effort is required. We should not alone strive for the attainment of practical results, but continued attempts should be made to ascertain all of the theoretical factors which may contribute to chemotherapeutic action.

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INSULIN

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PART I. PHYSIOLOGY

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As an outcome of the well-known discovery of von Mering and Minkowski (1) (1889) that pancreatectomy results in complete diabetes, it came generally to be assumed that the pancreas must produce an internal secretion, or hormone, having the function of controlling the metabolism of the carbohydrates in the animal body. The observations (Diamare (2), Laguesse (3), etc.) that the peculiar islets of cells that had previously been described by Langerhans (4)

(1869) occur in, or are associated with, the pancreas in all vertebrates, coupled with the demonstration by Ssobolew (5) and Schulze (6) (1900) that they do not undergo destruction after ligation of the ducts or in grafts of the gland—in neither of which conditions diabetes occurs—indicated that this anti-diabetic hormone must be derived from the islets. The name insulin was, therefore, suggested for it by Sir E. Sharpey Schafer (7) (1916).

Attempts to isolate insulin in extracts of pancreas were at first of no avail, although they almost succeeded in 1908. In this year Zuelzer (8) prepared from the pancreas of recently fed animals an alcoholic extract which on intravenous injection was capable of greatly diminishing, in several animals, the glycosuria resulting from injections of epinephrin and (in 1 animal) from pancreatectomy. For an hour or so prior to killing the animals, from whose pancreas the extract was to be prepared, the veins of the gland were tied so as to cause congestion, the object being, apparently, that the hormone might accumulate in it. The extracts contained considerable amounts of protein. Zuelzer subsequently administered portions of his extract to 8 patients suffering from diabetes mellitus and found that the glycosuria was greatly diminished in intensity and that the ketone bodies usually disappeared from the urine. Several of the patients improved in their general condition but as a rule considerable degrees of fever temporarily followed the injections. The extract was usually given intravenously and it was observed that its effects on the glycosuria did not develop until the second or third day following the injection and that they persisted for several days. Forschbach (9) confirmed Zuelzer's findings on depancreatized dogs but considered that the alleviation of the symptoms in diabetic patients was due to the febrile reaction which followed the injections, rather than to a specific action of an antidiabetic hormone. Further attempts to use Zuelzer's extracts in the treatment of diabetes were abandoned.

The next attempt to demonstrate the presence of insulin was that of Knowlton and Starling (10) in 1911. The extracts were made with weak acid, following the procedure used for preparing secretin, and they were tested by observing their effect on the rate of disappearance of sugar from the fluid perfused through the surviving

mammalian heart. Several of the experiments yielded positive results, but investigation of the behavior of the glycogen in the heart induced Starling (10), later, to consider it possible that a mistake in interpretation had been made, and further work with pancreatic extracts was abandoned by him. This research is, however, important because it pointed a new way to the testing of the extracts, and in 1916 Clark (11) succeeded in showing that the sugar consumption by the excised mammalian heart is definitely increased when the Locke's solution with which it is perfused is first of all passed through the blood vessels of the pancreas. The results of the investigation demonstrated beyond doubt that this gland produces an internal secretion which is capable of influencing the rate at which the heart removes sugar from the fluid perfused through it. Clark later showed that, although no change occurs in the percentage of sugar contained in the Locke's solution by perfusing it through the pancreas, as determined by the ordinary reduction methods, the sugar does become altered in some way as revealed by examination of the polarizing power of the solution. It cannot be said, however, that the changes observed were of sufficient magnitude to make this certain. Besides these researches, reference must also be made to those of Rennie and Fraser (12) (1907), Gley (13) (1905), E. L. Scott (14) (1911), J. R. Murlin and Kramer (15) (1913), I. Kleiner (16) (1919) and Paulesco (17) (1921). All of these investigators, using various methods, both for preparing the extracts and for testing their possible antidiabetic properties, were able to secure results which, though unconvincing, were, nevertheless, of such a nature as to keep alive the hope that some day it would be possible to obtain satisfactory pancreatic extracts containing insulin. Rennie and Fraser took advantage of the fact first discovered by Diamare, and further developed by Laguesse and by Rennie, that the islet tissue in certain of the bony fishes (Teleostei) is isolated from the main zymogenous tissue of the pancreas and exists as relatively large glands situated in the mesentery, usually near the gall bladder. The administration of extracts of these "principal islets," by mouth, did not definitely improve the clinical condition in diabetes, though, in one case, subcutaneous injection did appear to reduce the glycosuria and to improve the general condition of the patient. This method of

administration was not further attempted because toxic symptoms followed the injections. E. L. Scott, acting upon Ssobolew's suggestion, prepared extracts from the pancreas after attempting to cause atrophy of the acinous tissue by ligation of the ducts, but he discontinued the observations because he found that complete atrophy of the acinous tissue did not occur. He then tried alcoholic and acidulated watery extracts of the intact gland, the object in all his methods being to circumvent the destructive action of the digestive enzymes. He found, particularly with acidulated watery extracts, that intravenous injection into diabetic dogs reduced the glycosuria temporarily and seemed to improve the general condition of the animals. A slight rise in body temperature was considered by Scott to be a possible factor in accounting for the decreased glycosuria. Murlin and Kramer prepared extracts from the pancreas (dogs or cattle) either with acidified Ringer's solution, or with weak alkali. They tested the immediate and remote effects following subcutaneous injection of the extracts into diabetic dogs and they observed the behavior of the respiratory quotient along with the degree of glycosuria—and, in some cases, of hyperglycemia—to ascertain whether or not an antidiabetic hormone was present. Sometimes these workers combined the extracts of pancreas with extracts of duodenum, thinking that the secretion present in the latter might activate the antidiabetic hormone. Many of the results show evidence of the presence of insulin in the extracts, particularly certain of those in which the respiratory quotient was examined. Unfortunately, the observers failed to interpret their results correctly, being confused by the possibility that the alkali present in the extracts might in itself account for most of the beneficial results. Their conclusion "after some three year's intermittent occupation with the problem" is "that in the totally depancreatized dog in good condition, i.e., not moribund, the alkali alone is almost always without effect on the respiratory metabolism. . . . With the partially operated animal, however, the result is striking and immediate. . . . We have obtained some evidence also, that pancreatic material . . . is, when administered with alkali, of distinct benefit to the diabetic organism."

In collaboration with Clough, Stokes, Gibbs and Stone, Murlin (18) has more recently repeated Clark's experiments of perfusion of the blood vessels of the pancreas, using a variety of perfusates, such as weak alkaline, or acid or glucose solutions. Considerable quantities of insulin were found to pass into the perfusion fluids, particularly when the latter were acid in reaction. The antidiabetic properties were tested by various methods and in one case the life of a depancreatized dog was prolonged to thirty days by injecting perfusates in seven doses. There is nothing of practical importance in the results since it is to be expected that such perfusion of a half dead gland would lead to the extraction of some of the antidiabetic hormone.

Kleiner, continuing work which he had previously done with Meltzer, studied the influence of pancreatic extracts on the degree and duration of the hyperglycemia which follows injection of glucose into depancreatized dogs. The extracts were made with water and were unfiltered. After being diluted with isotonic saline they were administered intravenously at a slow rate. Decided reduction in the hyperglycemia was demonstrated to occur and it was shown by suitable controls that dilution of the blood was not responsible for the result.

There could be little doubt as to the existence of insulin in the pancreas, the problem—and for practical reasons a most important one—was to devise means for extracting it in a condition suitable for continued administration to diabetic patients. With this object in view F. G. Banting (19) undertook to prepare extracts from pancreas that had been caused to undergo partial degeneration by ligation of the ducts. Since it was known that the acinous tissue, from which the digestive ferments are derived, undergoes degeneration more rapidly than the islets which are presumably the source of insulin, it was hoped that in this way active extracts could be obtained. With the assistance of C. H. Best, the antidiabetic properties of the extracts were tested by injecting them into depancreatized dogs and observing the effects produced on the hyperglycemia and glycosuria. The results of a typical experiment are shown in figure 1. The animal was depancreatized on August 11 (1921) and extract, made by extracting the degenerated pancreas (ten weeks

after ligation of the ducts) with ice cold Ringer's solution, was given intravenously six hours afterwards. This prevented the blood sugar from rising above 0.20 per cent until 10:00 p.m. on August 12 when it reached 0.3 per cent. The amounts of extract injected

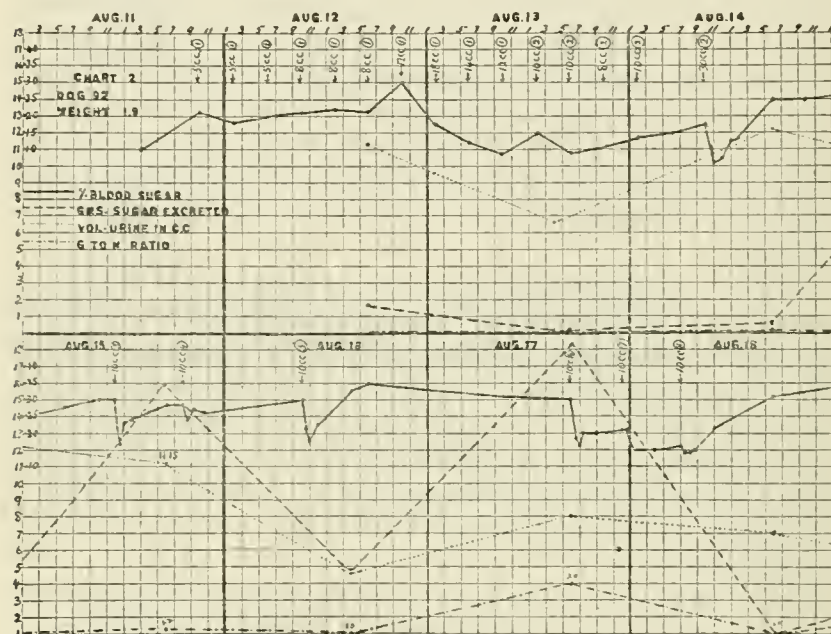


FIG. 1. CURVES OF PERCENTAGE OF BLOOD SUGAR, ETC., IN DEPANCREATIZED DOGS, AS AFFECTED BY INJECTIONS OF EXTRACTS OF PARTIALLY DEGENERATED PANCREAS

(1) Degenerated pancreas, dog 394. (2) Degenerated pancreas, dog 390. (3) Degenerated pancreas in 0.1 per cent HCl. (4) Degenerated pancreas in 0.10 per cent NaOH. (5) Degenerated pancreas in 0.1 per cent HCl. (6) Whole gland extract, fresh, cold. (7) Whole gland extract in 0.1 per cent HCl. (8) Whole gland extract in 0.1 per cent NaOH. Dog died August 30. The first column of figures along ordinates refers to all the curves except that of the percentages of blood sugar which are given in figures of the second column. In the case of "volume of urine" the figures are to be multiplied by 100.

were then increased with the result that the blood sugar declined, reaching a little below 0.10 per cent by noon on August 13. After this, similarly prepared extract from another degenerated gland was used, and on August 14 an isolated, large injection resulted in a fall of blood sugar from 0.175 per cent to nearly 0.09 per cent.

Meanwhile very little sugar was being excreted by the urine, but as the effect of the last injection of extract wore off the amount rose rapidly and on August 15 reached 16 grams, the blood sugar now standing at 0.30 per cent. Injections of extract prepared by weak acid from degenerated pancreas on August 15 and 16 caused prompt reductions in blood sugar, and the urinary sugar again declined to about 5 grams. There could be no doubt that the extracts of degenerated pancreas had reduced the severity of the diabetic symptoms, and on August 17 and 18 extracts from normal (dog) pancreas were tried with similar beneficial results, as judged by both the hyperglycemia and the glycosuria. The observations were continued on this animal until August 22, one of the extracts used with successful results being prepared from a pancreas after exhaustion of the acini by continued injections with secretin. After this date, as a result of discontinuance of the injections the blood sugar rose to between 0.30 and 0.35 per cent, at which it remained until August 26, the daily urinary sugar varying between 6 and 13 grams. The animal died on August 30 and at autopsy no trace of pancreas was found by naked eye examination. In other experiments, in which extracts prepared by similar methods from other tissues than the pancreas were used, it was shown that insulin is almost, if not exclusively, derived from this gland. Administration *per rectum* was without effect. In certain of the experiments glucose solutions were injected and it was found that when this was accompanied by the extract, decidedly less than the expected amount of glucose reappeared in the urine. Thus, in one experiment without extract the excretion of glucose during four hours following the injection of 10 grams was 9.94 grams, whereas on the next day when extract was also given, it was only 4.4 grams. Hemoglobin determinations showed that the changes in the percentage of blood sugar were not due to dilution of the blood.

Observations of a similar type were made on 9 other depancreatized dogs, partly in order to make certain that no fortuitous circumstances could be held responsible for the results, and partly in the endeavor to obtain more satisfactory figures for the sugar excretion by the urine.

No attempt was made at this stage to investigate the possible effects of the extracts on other symptoms of diabetes, such as glycogen

formation or ketosis, since, with the limited quantities of extract that were then available, it was considered more important to devote attention first of all to finding methods for the preparation of larger supplies, using the behavior of the blood and urinary sugar of diabetic dogs to determine their antidiabetic potency.

As the next step towards obtaining larger amounts of extract the pancreas of fetal calves of under four months development was used by Banting and Best (22), since it had been shown by Ibrahim (20) that at this stage the acini of the pancreas are insufficiently developed to produce active proteolytic enzymes, whereas the islets are plentiful and are apparently capable of supplying sufficient internal secretion of insulin to minimize greatly the severity of glycosuria in the mother (Carlson and Drennan (21)). These extracts were made partly with Ringer's solution and partly with alcohol. In the latter procedure, the alcohol was removed by evaporation in a current of warmed air and the residue redissolved in Ringer's solution. Intravenous or subcutaneous injections into depancreatized dogs reduced the hyperglycemia and glycosuria. This result encouraged Banting and Best (22) to see whether potent extracts could be obtained from the pancreas of full grown cattle by extraction with equal volumes of 95 per cent alcohol made slightly acid with HCl. This extractive was used with the object of minimizing the destructive action of the proteolytic enzymes, and its possible value in this connection, previously recognized by Zuelzer and E. L. Scott, had been in mind from the very start of the investigations. After the removal of the alcohol by warmed air and of excess of fat by toluene, the extracts were found to possess strong antidiabetic properties, and it was now possible to show without doubt that by continuous injections a great improvement occurred in the general condition of the animals, one of which lived for seventy days, when it was killed by chloroform. On gross examination no trace of the pancreas could be found but serial sections of the duodenum, made by Dr. W. L. Robinson, revealed the presence in the submucous coat, near the entry of the main pancreatic duct, of a small nodule in which, however, no islets could be seen.

An endeavor was now made to purify the alcoholic extracts of adult pancreas sufficiently for trial on a diabetic patient. The alcohol

was removed by warmed air or *in vacuo* at a low temperature, the excess of fat extracted by toluene and the watery residue, now reduced to one-fifth the original volume, was passed through a Berkefeld filter. The resulting extract, injected into a diabetic patient (a boy aged fourteen years), reduced the blood sugar by a little over 25 per cent, and somewhat diminished the glycosuria, but "owing to the high percentage of protein . . . sterile abscesses formed in a few instances at the site of injection." Banting and Best (23) observed that the potency of these extracts is destroyed by heat and by digestion with trypsin and that the active principle is insoluble in 95 per cent alcohol.

Before further attempts could be made to investigate the possible therapeutic value of the extracts it was necessary to remove from them the irritating substances responsible for abscess formation, and to demonstrate in diabetic dogs that not only are the hyperglycemia and glycosuria reduced by their action, but the other diabetic symptoms as well. At the same time it was considered important to see whether other forms of experimental hyperglycemia, such as that caused in rabbits by piqûre or epinephrin, would be affected by the extracts. The problem of purification of the extract was entrusted to J. B. Collip (24) who, as a first step in his work, injected some of the crude extract into normal rabbits and found the blood sugar to become reduced. This furnished him with a method for testing the potency of the various precipitates and filtrates which were produced in the crude alcoholic extracts by various strengths of alcohol. He finally found that the active principle remained in solution up to an alcohol percentage of about 92 and that by using percentages somewhat below this, much of the protein could be precipitated from the extracts. His finally evolved method was as follows:

To a small volume of 95 per cent ethyl alcohol freshly minced pancreas was added in equal amount. The mixture was allowed to stand for a few hours with occasional shaking. It was then strained through cheese cloth and the liquid portion at once filtered. The filtrate was treated with two volumes of 95 per cent ethyl alcohol. It was found by this treatment that the major part of the protein was removed while the active principle remained in alcoholic solution. After allowing some hours for the protein precipitation to be effected the mixture was filtered and the filtrate con-

centrated to small bulk by distillation *in vacuo* at a low temperature (18° to 30°C.). The lipoid substances were then removed by twice extracting with sulphuric ether in a separating funnel and the watery solution returned to the vacuum still, where it was further concentrated till it was of a pasty consistency after which 80 per cent ethyl alcohol was added and the mixture centrifuged. After centrifuging, four distinct layers were manifested in the tube. The uppermost was perfectly clear and consisted of alcohol holding all the active principle in solution. Below this, in order, were a flocculent layer of protein, a second clear watery layer saturated with salt and a lowermost layer consisting of crystals of salt. The alcohol layer was removed by means of a pipette and was at once delivered into several volumes of 95 per cent alcohol, or better, of absolute alcohol. It was found that this final treatment with alcohol of high grade caused the precipitation of the active principle along with adherent substances. Some hours after this final precipitation the precipitate was caught on a Buchner funnel, dissolved in distilled water and then concentrated to the desired degree by use of the vacuum still. It was finally passed through a Berkefeld filter.

Various *methods* have been devised to purify further the insulin prepared by Collip's method and as a result preparations giving only a faint biuret test have been obtained. The best known methods are those of Doisy, Somogyi and Shaffer (25), and of Dudley (26). In the former, the acidified alcoholic extract of pancreas, after removal of the alcohol, is mixed in faintly acid reaction with ammonium sulphate in the proportion of 40 grams to each 100 cc. of solution. After standing in a separating funnel the precipitate rises to the surface so that the underlying fluid can readily be run off. When all possible liquid has been removed the precipitate is shaken with alcohol in such proportion as to give about 70 to 75 per cent, and the mixture is centrifuged. A clear alcoholic solution separates which is pipetted off and the concentration of alcohol in it raised to about 90 per cent, the reaction being carefully adjusted to a pH of between 5 and 6. After standing some hours, the precipitate which forms is collected and redissolved in water containing traces of acid (or alkali). Precipitation is again effected by bringing pH back to between 5 and 6, and this process of precipitation and resolution is repeated several times, the final precipitate being collected on a filter and dried in a vacuum desiccator. It will be seen that this method depends mainly on precipitation at the isoelectric point.

It is one which is being used successfully on a large scale and it yields insulin which is practically free from toxic or irritating qualities and and possesses remarkable keeping qualities. Its discoverers have generously allowed it to be used for large scale production.

In the Dudley process crude insulin prepared by Collip's process is dissolved in a small quantity of water and the solution centrifuged to free it from insoluble material. The clear supernatant solution is diluted with water to bring the concentration of insulin to about 1.5 per cent. An equal volume of a saturated watery solution of picric acid is added and the material, contained in a tall vessel, is allowed to stand some days so that the precipitate may settle. The clear supernatant fluid is decanted and the precipitate dissolved in a minimum of water containing weak Na_2CO_3 . If all does not dissolve the solution is centrifuged. Acid is added to the clear solution in an amount sufficient to neutralize the Na_2CO_3 as a result of which the insulin picrate is reprecipitated. To ensure complete reprecipitation one-half the volume of saturated picric acid solution is added to the solution. After standing some days the precipitate is collected on a Buchner funnel and thoroughly washed with weak picric acid solution (5 cc. saturated picric acid in 100 cc. water) after which it is transferred to a beaker and stirred with a solution of HCl in 75 per cent alcohol in the proportion of about 15 cc. per gram of moist picrate taken (24 cc. 3 *N* HCl in 75 cc. absolute alcohol). "The picrate after first forming thick dark brown oily drops dissolves in this reagent yielding a somewhat turbid dark yellow liquid." About ten volumes of acetone are then added to precipitate the hydrochloride which is collected on a filter, washed with acetone, and finally, with dry ether. It is then dried in a vacuum desiccator.

In the preparation there are two stages during which loss of insulin is liable to occur; one of these is prior to placing the minced gland in acidified alcohol and the other, in the watery extract after removal of the alcohol. In order to minimize the losses in the early stages Krogh and Widmark (27) place the pancreas, after removal from the animal, in a freezing mixture of ice and 2 per cent saline solution in which it is brought to the laboratory. It is then frozen solid and the blocks of ice cut by rapidly revolving knives into thin shavings which are collected in acidified alcohol. After extraction and removal of

the alcohol, the extract is tested as to stability of insulin by roughly assaying it before and after incubation for twenty-four hours in neutral reaction. If it is found to be satisfactorily stable the insulin is further purified by the ammonium sulphate method of Doisy, etc. The yields by this method are remarkably high, probably because of the great care that is taken to chill the pancreas to freezing point as early as possible after killing and to break up thoroughly the frozen gland so that extraction is thorough. We have frequently noted when working with the principal islets of bony fishes (cod, angler fish, sculpin, etc.) that the yields of insulin are greatly increased when the glands are frozen solid immediately after removal and we have attributed this, not only to the inhibition of proteolytic digestion, but also to the tearing up of the islet cells by the large ice crystals which form under these conditions.

Brailsford Robertson and Anderson (28) employ sodium sulphate to remove water from the initial alcoholic extracts, thus effecting considerable saving in alcohol. This action depends on the fact that exsiccated Na_2SO_4 when mixed with water forms the crystalline sulphate, $\text{Na}_2\text{SO}_4 \cdot 10\text{H}_2\text{O}$, and in the process absorbs 1.27 times its weight of water. Thus, by adding to the 50 per cent (initial) alcoholic extract enough Na_2SO_4 to remove four-fifths of the water present, the concentration of alcohol is raised to 80 per cent at which protein fractions not containing insulin are precipitated. Much less alcohol has, therefore, to be used and the sodium sulphate can be readily recovered for use again by igniting it.

Besides the mammalian pancreas the principal islets of bony fishes may also be used for preparing insulin. Since the yields from this source, per gram of tissue, are many times greater than from pancreas, the process of manufacture is incomparably simpler and less expensive and the final product is of a high degree of purity. It will be remembered that Diamare (2) originally suggested the anti-diabetic function of these glands (in 1899) and that Rennie and Fraser (12) in 1906 almost succeeded in demonstrating the presence of insulin in them. It was not until 1922, however, that final proof of the presence of insulin in the islets was demonstrated by Macleod, mainly in the angler or devil fish (*Lophius piscatorius*) and in the sculpin (*Myoxocephalus*) and more recently N. A. McCormick and E. C.

Noble have found it to be obtainable in very large amount from the islets of the cod (*Gadus*), pollock (*Pollachius*) and halibut (*Hippoglossus*). The islets in these very common fishes are easily recognizable and can be removed without difficulty and since insulin, sufficiently pure for clinical use, can be prepared by extracting them with a relatively small amount of alcohol (enough to give 60 per cent in the mixture) removing the alcohol by vacuum distillation or by warmed air, heating the watery extract to about 80°C. extracting with ether and then applying Dudley's process of purification, this should prove a useful source of supply, at least in maritime countries.

When larger quantities of purified insulin became available a thorough clinical trial of its therapeutic value was undertaken (cf. Banting and Best, Collip, Campbell and Fletcher (30)), and the results of these will be found on page 239. The present part of the article is restricted to the experimental aspects of the work in so far as these bear directly on the clinical use of insulin. These will be considered in two categories: (1) further observations on depancreatized dogs; and (2) observations on normal animals and on other forms of hyperglycemia, particularly with reference to the assay of insulin.

FURTHER OBSERVATIONS ON DEPANCREATIZED DOGS

Glycogen content of the liver and the heart

Various workers have confirmed the observation, first made by Minkowski (1), that no glycogen, or only traces, can be found in the liver of depancreatized dogs, even after the animals are fed large quantities of glucose. Minkowski thought, however, that some was deposited with levulose. Cruickshank (31), while confirming these conclusions with regard to glucose, considers that Minkowski's positive results with levulose may have been due to incomplete pancreatectomy, since he himself failed to detect glycogen after feeding with this sugar. We can contribute two observations bearing on this question (32). In a dog which lived for seven days following pancreatectomy, cane sugar was fed in large amounts during the last three days of life and 1.32 per cent of glycogen was found in the liver; in another, which lived for eleven days, only 0.046 per cent was found. In neither

of these animals did the cane sugar cause any change in R.Q. Since serial microscopic sections of the duodenum were not made, it is possible that a trace of pancreatic tissue may account for the glycogen formation in the one animal.

In any case we can be certain that even by feeding with cane sugar very little glycogen becomes deposited in the liver of depancreatized dogs, which contrasts very strikingly with the result which is obtained when insulin is given at the same time as the sugar. The first observation in which this was done was made on a dog (15 kgm. weight) that

TABLE 1

DATE	DAYS DEPAN- CREATIZED	DAYS DURING WHICH INSULIN AND SUGAR WERE GIVEN	PER CENT GLYCOGEN IN LIVER
February 21 (1922).....	7	5 days	12.58
January 14.....	4	Less than 1 day	2.70
April 28.....	3	2 days	11.40
May 2.....	5	1 day	4.80

TABLE 2

DATE	DAYS DEPAN- CREATIZED	GLYCOGEN CONTENT	
		Liver	Heart
		<i>per cent</i>	<i>per cent</i>
April 24 (1923).....	5	0.09	0.47
May 1*.....	4	Trace	0.38
May 4.....	5	Trace	0.12
May 5.....	3	0.06	0.61
May 7.....	4	0.06	0.33
May 11.....	3	0.07	0.65
May 14.....	4	0.03	0.66

had been depancreatized during nine days (December 14 to 22, 1921), no insulin being given until two days before death. Although only the crude extract was available for administration, it lowered the blood sugar from 0.309 per cent to 0.051 per cent, in spite of the fact that, at the same time, large amounts of cane sugar were fed to the animal, which it took greedily. Analysis of the liver by Pflüger's process was found (by J. B. C.) to contain an unprecedented amount of glycogen (over 20 per cent). Other observations of a similar nature are shown in table 1 (32). For comparison, results obtained on depan-

creatized dogs to which insulin was not given are shown in table 2 (McCormick (33)). In these, except the one asterisked, the animals were under ether anesthesia for at least one hour before being killed.

In several of the observations glycogen was also determined in the muscles and the heart. Nothing conclusive can be made out from the analyses on the skeletal muscles but there is evidence that insulin causes reduction in the amount of glycogen in the heart in which, as histological examination by previous workers had shown, unusually large deposits of this substance occur, both in diabetic patients and in depancreatized dogs. Cruickshank (31) found by analysis (on 16 depancreatized dogs) the average percentage of glycogen to be 0.7 compared with 0.5 per cent in normal animals (6 observations). We found 0.79 to 0.92 in different portions of the heart of one depancreatized animal fed sugar but not given insulin, and 0.98 per cent in that of another similar animal. In 4 depancreatized animals to which insulin, as well as sugar, was given the values were 0.725, 0.600, 0.570 and 0.296 per cent respectively. It must be pointed out, however, that although the glycogen content of the heart is often high in depancreatized dogs this is not invariably so. Out of the 7 animals reported above, it will be seen that high percentages occurred in only 3, and that it was very low in 1 case (0.12 per cent). The ether anesthesia may account for the unexpectedly low percentages of glycogen found in this series of observations.

Fatty acid content of the liver and blood

It is well known that in diabetes the percentage of fats rises both in the liver and the blood. In the former, as the glycogen disappears fats accumulate and may reach a very high percentage (over 20 per cent); in the latter, especially in the terminal stages of the disease, percentages of over one are not uncommon. The effect of insulin on the total fatty acids determined by Leathes' modification of the Kumagawa-Sato method is shown in table 3.

In two of the observations, Nos. 53 and 54, the results are not conclusive since such excessive doses of insulin were administered that death from hypoglycemia occurred within twelve to eighteen hours. There is sufficient evidence from the other results, however, to show that insulin reduces the fat content of both liver and blood. The

effect on the blood fat appears to be very rapidly developed (within a few hours) which is of interest on account of the similar prompt effects of insulin on the excretion of the ketone bodies. As far as can be judged from the available analyses the effect on the liver fat is not so prompt, which indicates, when taken in conjunction with the be-

TABLE 3
Total fatty acids in depancreatized dogs, with or without insulin

NUMBER OF ANIMAL OR DATE OF OBSERVATION	TOTAL FATTY ACIDS		REMARKS
	Liver*	Blood†	
48	12.25		} Fed sugar but insulin not given
50	14.10	1.21	
51	9.90	1.12	
52	7.428	0.30	} Sugar + insulin 1 day Sugar + insulin 2 days Sugar + insulin 1 day Sugar + overdose insulin‡ Sugar + overdose insulin
55	2.190	0.53	
56	4.41		
53	10.28		
54	26.36	0.37	
April 24	{ 25.3 23.9	{ 1.07 1.08	} Not fed sugar nor given insulin
April 27	12.80	1.02	
May 1	12.75	1.13	
May 4	33	1.58	} The number of days since pancreatectomy will be seen by comparing dates given in previous table
May 5	8.8	0.72	
May 7	23.1	0.74	
May 11	15.6	{ 0.85 0.91	
May 14	26.24	{ 0.76 0.79	

* Leathes' method.

† Bloor's method.

‡ These animals died within a few hours after insulin.

havior of the blood fat, that the removal of fat from the liver must proceed at a pace which is conditioned upon the rate at which it can be deposited in the fat depôts or converted into other lipid substances. The remarkable effect of insulin on lipemia in diabetes mellitus has been repeatedly confirmed in the clinic.

Excretion of ketone bodies in the urine

The oxidation of fats in the animal takes place at the β -carbon atom (i.e., the C atom two places removed from the COOH group) and the process proceeds smoothly, step by step, until all the methyl radicles have been consumed. In diabetes this process proceeds smoothly only so far, leaving a residue still containing three methyl radicles, only one of which, in the β -position, undergoes partial oxidation forming a series of substances known as the ketone bodies β -oxybutyric acid, aceto-acetic acid and acetone ($\text{CH}_3\text{CHOH}\cdot\text{CH}_2\cdot\text{COOH}$; $\text{CH}_3\cdot\text{CO}\cdot\text{CH}_2\cdot\text{COOH}$; $\text{CH}_3\cdot\text{CO}\cdot\text{CH}_3$). These bodies have serious

TABLE 4
Excretion of sugar and ketone bodies in depancreatized dogs

NUMBER	DATE	INSULIN GIVEN	TOTAL URINE	TOTAL DEX- TROSE EXCRE- TION	TOTAL ACE- TONE BODIES	REMARKS
			cc.	grams	mgm.	
11	January 6	No	1,000	29.7	100	
	January 7, a.m.	No	375	28.4	187	Blood sugar 0.351 per cent
	January 7, p.m.	Yes				Blood sugar 0.085 per cent
	January 8	No	425	4.25	None	
	January 9	No	325	9.95	None	
	January 10	No	370	9.6	None	
	January 11	No	275	25.2	34	
	January 12	No	325	25.4	55	
	January 13, a.m.	No	750	18.0	114	
	January 13, p.m.	Yes				
	January 14	Yes	600	8.0	None	

poisonous effects on the animal (coma) due partly to their acidic properties. It was considered important to observe their behavior in depancreatized dogs given insulin. The total ketone bodies were measured in the daily urine of three depancreatized dogs (by J. B. C.) using the Van Slyke method. In one of them the daily excretion steadily rose during seven days following pancreatectomy without insulin, from 75 to 210 mgm. the daily sugar excretion on a meat diet varying between 24 and 36 gms. On the seventh and eighth days massive doses of (crude) insulin were given along with sugar, and both the ketone bodies and the sugar disappeared entirely from the urine.

(This is the same animal on which the first glycogen determination was made, see page 208.)

In another animal the results given in table 4 were obtained.

These results show clearly the prompt effect of insulin in removing ketonuria and it has not been considered necessary to repeat the observations on depancreatized dogs, since this can be more conveniently done in human diabetics. The clinical observations (34) have demonstrated a similar reduction both in the urine and the blood. The concentration of ketone bodies in the blood decreases somewhat more slowly than the sugar and the decrease is accompanied by a rise in the CO_2 -combining power. In the urine of diabetic patients the ketone bodies disappear later than the sugar, following the administration of insulin. These observations indicate the necessity of a very careful comparison of the relative speeds with which insulin clears up lipemia, ketonuria and glycosuria in diabetes and the relation of the time of disappearance of these symptoms to changes in blood sugar and the respiratory quotient. By such comparisons it is possible that light may be thrown on the mechanism of the action of insulin.

Behavior of the respiratory quotient

It is generally recognized that the most satisfactory evidence of the utilization of carbohydrate in the animal body is afforded by the behavior of the respiratory quotient, i.e., the ratio between the volume of CO_2 expired and O_2 absorbed. In the normal animal this quotient approaches unity in proportion as carbohydrates replace fats and proteins in the total metabolism; thus, when sugar is given to animals that are starving or living on a fat and protein diet the quotient promptly rises. In the completely diabetic animal, on the other hand, whether this condition be brought about by removal of the pancreas, by administration of phloridzin or by disease, the quotient remains at the level of about 0.7 or below it (which is characteristic of the metabolism of a mixture of fat and protein) even when large amounts of carbohydrate are ingested.

The expired air of diabetic dogs was collected in a Tissot spirometer and analyzed by the usual technique (by Hepburn and Best (35)). The procedure followed was to determine R.Q. for periods

preceding and following the administration, by mouth or subcutaneously, of cane sugar or glucose, and then to repeat the observation giving insulin, as well as the sugar. Since previous investigators (Murlin (15)), Verzar (36), etc.) had shown that the power to oxidize carbohydrate, as revealed by the behavior of R.Q., does not entirely disappear until about four days following pancreatectomy the observations were not made until after this period. The following is a typical result:

DATE	CONDITION	R.Q.
<i>1922</i>		
January 20	Normal dog, starved 24 hours	0.85-0.86
	35 minutes. After 30 grams sucrose	1.0
	1 hour 25 minutes later	1.0
January 21	Pancreas removed	
January 23	No sugar or insulin	0.63
	1½ hours after 20 grams sucrose	0.77
January 24	30 minutes after 25 grams sucrose and insulin	0.86
	1½ hours later	0.90
	3 to 3½ hours later	0.77
January 25	No sugar or insulin	0.68
	various periods after 20 grams sucrose	0.82, 0.82, 0.85
January 26	No sugar or insulin	0.68-0.72
	1 hour after 20 grams sucrose	0.81
	5 hours later and 40 minutes after insulin	0.90

In another animal, six days after pancreatectomy, sugar and insulin raised the quotient to 0.95 previous observations with sucrose alone succeeding in raising it only from 0.63 to 0.70. The results show that in depancreatized animals insulin and sucrose cause a greater rise in R.Q. than occurs with the same quantity of sucrose alone. In both the earlier and the more recent observations the effect of insulin in raising the respiratory quotient in depancreatized dogs has been observed to persist for about two days following the injection. This shows that the action of insulin on diabetic dogs can be detected for a longer time, following the injection, when the behavior of the respira-

tory quotient is used to indicate it, than when the level of the fasting blood sugar is used (cf. fig. 1).

Similar results have been obtained by S. U. Page (36c) in phloridzin diabetes. Thus, in a dog which was injected daily with 2 grams of this drug and was on a constant meat diet, the R.Q. remained at about 0.7 and was unaffected by giving 40 grams of sucrose subcutaneously. When insulin was given as well as sugar, however, R.Q. rose, as shown in table 5.

In the observations made during July the daily diet was 400 grams meat and in those made during September it was 600 grams. The excretion of glucose in the urine collected during twenty-four hour periods was also measured but the results did not demonstrate a marked decrease as a result of insulin. The excretion of ketone bodies was diminished by the injections. More recently M. Ringer (36a) and Nash (36b) have made similar observations under more carefully controlled conditions. Ringer found that when 40 grams glucose were given along with insulin (20 units) 12.2 grams failed to reappear in the urine and R.Q. rose from 0.71 to 0.75 indicating that about 6 grams could be accounted for by increased oxidation. On another occasion R.Q. rose to 0.84. The experiments on the combined effects of insulin and phloridzin are difficult to conduct because hypoglycemic symptoms are very readily induced. It is on this account that Page was unable to obtain satisfactory results on the influence of insulin on the excretion of sugar in the urine. Ringer suggests that the effect of insulin on phloridzin diabetes might be used as a basis for its assay. We do not believe that the method is likely to prove one of practical value.

Reviewing these results as a whole it is clear that insulin plays an essential rôle in the metabolism of carbohydrates, as well as of fats. Both types of metabolism proceed in the normal animal through an extended series of chemical changes. In the case of carbohydrates, glucose, after its absorption, becomes either polymerized to glycogen and stored in this form or gradually broken down into simpler products which are finally oxidized. It is possible that glycogen formation is a necessary preliminary step to this catabolism. What the intermediary products may be is unknown, but there are reasons for believing that they are numerous and that they exist in a certain balance with one

TABLE 5
Effect of insulin on R.Q. in phloridzin diabetes

DATE	TIME	CONDITION	R.Q.
1922			
July 5		Phloridzin, 2 grams daily, started.	
July 10	10:30 a.m.		0.70
	12:00 n.	40 grams sucrose	
	1:15 p.m.		0.71
July 11	10:15 a.m.		0.67
	11:50 a.m.	insulin	
	12:50 p.m.	40 gms. sucrose	
	2:15 p.m.		0.91
July 12	10:30 a.m.		0.72
	1:00 p.m.	40 gms. sucrose	
	2:15 p.m.		0.83
September 2		Phloridzin, 2 grams daily, started.	
September 8	10:10 a.m.		0.71
	12:10 p.m.	40 grams sucrose	
	1:25 p.m.		0.76
September 9	9:25 a.m.		0.72
	10:50 a.m.	insulin	
	11:00 a.m.		0.70
	1:15 p.m.		0.92
September 11	10:15 a.m.		0.79
	1:15 p.m.	40 grams sucrose	
	2:30 p.m.		0.84
September 13	9:30 a.m.		0.74
	10:50 a.m.	insulin	
	12:55 p.m.	40 grams sucrose	
	1:10 p.m.		0.88

another. In such a case it is evident that the failure of *one step* in the break-down process would interfere with the entire process.

It has been suggested by Winter and Smith (37) that the first step is the conversion of glucose into a more reactive form and that insulin

is necessary for its occurrence. Chemically considered ordinary glucose is an equilibrated mixture of two varieties, designated α and β , characterized by differences in their power to rotate the plane of polarized light, and in both of which the carbon atoms are joined in a ring formation which makes them relatively stable. In the more reactive form, designated γ -glucose the rotary power is different (rotates to the left instead of to the right) and it is believed that the molecule has become less stable because of a shifting in the position of the ring (Irvine (38), etc). These varieties cannot be identified in animal fluids by the ordinary reduction tests, but only by their different rotatory powers. Using this method, Winter and Smith have claimed that γ -glucose can be detected in normal blood (of laboratory animals and men) but not in diabetic blood, in which however, it appears when insulin is given. Too much weight must not be placed on these results, however, partly because the differences (in rotatory power) observed are very small, partly because very complicated processes have to be employed to remove the proteins from the blood and partly because the results have not so far been confirmed by other workers. Using the same technique G. S. Eadie (39) has indeed obtained polarimetric readings similar to some of those reported by Winter and Smith for normal blood, but on the contrary he has also obtained others of an exactly opposite character. Winter and Smith publish very few protocols so that it is difficult to appraise their work correctly. One serious objection to their conclusions is that they do not adequately rule out other possible explanations for their results. Prior to the work of Winter and Smith, Hewitt and Pryde (40) had stated that optical evidence of the formation of γ -glucose can be obtained when a solution of glucose is placed on a loop of the intestine for a short time. Stiven and Waymouth Reid (41) and more recently van Creveld (42) have shown, however, that such is not the case and the last mentioned worker has also been unable to confirm any of Winter and Smith's conclusions.

Apart from this no attempt has so far been made to determine the exact stage in carbohydrate metabolism for the consummation of which insulin is required. That both the formation of glycogen and the oxidation of glucose are interfered with in diabetes indicates that

a relationship must exist between the two processes and this can be explained most simply by supposing that glucose, before it proceeds to final oxidation, must pass through a glycogen stage for which insulin is necessary. There is no direct evidence at present to support this view but it is at least significant that after pancreatectomy, the respiratory quotient does not reach its final low level, of about 0.65, until all the glycogen has disappeared from the liver. It remains to be determined whether some glycogen is formed prior to the increase in respiratory quotient, when sugar and insulin are given to diabetic animals. Such an hypothesis would place glycogen in a more important position in metabolism than that of a mere storage form of carbohydrate. Its formation may be a necessary step in the metabolism of sugar and it is conceivable that it is so because the glucose which is formed by its hydrolysis in the body is of a different type from the α - β -glucose from which it was built up. An important objection to this hypothesis is that glycogen is a polymerized form of glucose and therefore a step further away than glucose itself from the break-down products which finally become oxidized. It is easier to imagine that insulin, perhaps in association with a co-enzyme, converts glucose into some active form which is then used to meet the demands of the tissues for carbohydrate, and that the remainder is polymerized into glycogen and stored until no more is being absorbed from the intestine, when the glycogen again breaks down to glucose. The persistence of glycogen in muscle and particularly in the heart after the liver has become glycogen-free, as in diabetes and acute starvation, would however, seem to indicate that it must be more than a mere storage material. Under these conditions it is formed at the expense of protein, and, it is pertinent to enquire why should it be so unless it is an essential step on the road to oxidation?

Turning now to fats, it is again significant that in diabetes the long series of changes through which they pass should be interfered with at both ends of the metabolic chain, as it were, and that insulin should not only immediately restore to normal the migration of fat between the liver and tissues but also rectify the faulty oxidation. It is usually stated that fats burn in the fire of carbohydrates, but the exact relationship between the metabolism of these two substances is not clearly

understood. Although much progress has recently been made towards furnishing an answer to this problem, it cannot be said that enough is known to make it possible to understand how insulin brings about these effects.

Some further facts concerning the effect of insulin on depancreatized dogs have more recently been elucidated largely through work by Frank N. Allan (43). These studies have had two main objects: (1) to determine the glucose equivalent of insulin and, (2) to ascertain how long a diabetic animal can be kept alive by continuous treatment with it.

The glucose equivalent of insulin means the number of grams of glucose which a given quantity of insulin can cause to be metabolized in a diabetic animal (depancreatized dog), and it is measured by comparing the glucose balance of one period during which a certain amount of insulin is given with that of another period during which the amount of insulin is changed. A method based on this principle has been used in the clinic but the results have been inconstant, due in part, no doubt, to the fact that the diabetic patient retains some remnant of islet tissue still capable of yielding an endogenous supply of insulin which is not constant from day to day.

The animals after pancreatectomy were fed daily with a liberal quantity of flesh (500 to 600 grams) as free from fat as possible, and 100 to 150 grams of cane sugar. This ration was given twice daily and insulin injected subcutaneously immediately before each feeding. The total daily glucose excretion was compared with the glucose intake (i.e., cane sugar $\times 1.053$ plus glucose from protein obtained by multiplying urinary nitrogen by 3.625). The difference between excretion and intake gives the amount of glucose metabolized in twenty-four hours. When this is determined for two periods during which different amounts of insulin are given it is easy to compare the glucose equivalents of the insulin. The following facts have been established: (1) The glucose equivalent of the same dose of insulin given on different days of equal glucose intake varies within a relatively narrow range. (2) It is relatively much larger (per unit of insulin) with small than with large doses of insulin. (3) It becomes greater (per unit) when the amount of ingested carbohydrate is increased. The first two conclusions are based on the following results:

Dog 1. Fed 600 grams meat and 100 grams sucrose.

INSULIN GIVEN PER DAY (CLINICAL UNITS)	GLUCOSE EQUIVALENTS ON DIFFERENT DAYS
20	5.8, 5.8
24	4.1, 4.0
32	3.5, 3.5, 3.6, 3.3, 3.7, 4.1
36	3.0, 3.1, 3.1
38	3.4
40	3.0, 3.3, 3.1, 3.0

The third conclusion is based on the following:

Insulin given daily 20 units

SUGAR GIVEN PER DAY	GLUCOSE EQUIVALENTS ON DIFFERENT DAYS
<i>gms.</i>	
50	3.5, 3.9
100	5.8, 5.8
150	8.4, 8.1, 6.9, 6.9

The interpretation of these results is that when the amount of insulin exceeds a certain optimum some of it does not have an opportunity of affecting the metabolism of glucose. This may, in part, be due to the fact that with larger amounts of insulin some is excreted before it can act on the sugar, but other factors must also come into play as is clear from the results obtained by keeping the insulin dosage constant but varying the sugar intake (third conclusion). The action of insulin on glucose metabolism no doubt proceeds according to the same general laws as govern enzyme action, namely that the amount of substrate which is acted on is not a linear function of the amount of enzyme but is relatively much greater when there is a large excess of substrate compared with the amount of enzyme. This latter condition is fulfilled when small amounts of insulin are given, or when, with larger amounts, the intake of sugar is increased. The same relationships between dosage and effect are seen in the pressor action of epinephrin and no doubt apply for drugs in general (cf. Murray Lyon (44)).

There are two practical applications of these results. One of them is of clinical importance, for it indicates that even very minute doses of insulin may be of great advantage to the diabetic patient in enabling him to metabolize much more glucose than he otherwise could.

Suppose, for example, that we give one unit of a certain preparation of insulin to a patient who is excreting a large amount of sugar, it will cause many more grams of sugar to disappear from the daily urine than if we give the same amount to a patient that is almost sugar-free. The other is pharmacological since it indicates a possible method for the accurate assay of insulin. Instead of expressing the dosage in terms of the power to lower the blood sugar of normal rabbits, upon which the present unit is based, it may be done in terms of glucose equivalents. But it is clear that we cannot state that one unit of insulin will cause so many grams of glucose to be metabolized, unless we know the glucose balance at the time the insulin is given.

The relation between insulin and glucose equivalents is such that when the logarithms of the amount of insulin given (expressed in rabbit units) are plotted against the logarithms of the glucose equivalents (the intake of glucose being constant), the points fall along a straight line, represented by the equation $\log g = 1.86 - 0.85 \log u$ and which may be expressed in the form $g(u)^{0.85} = 10^{1.86}$. The curve corresponding to this equation is given in figure 2 in which, also, the glucose equivalents (ordinates) actually found in two different animals for different doses of insulin (abscissae) are indicated by dots and crosses. Failure to recognize the fact that the glucose equivalent of each unit of insulin is greater with small doses than with large, explains in part at least the considerable variations obtained by clinical investigators. Taking the most dependable of those published by Wilder, Boothby (45), etc. the equivalent per unit was found to run between 0.9 and 3.1 grams with an average of 1.45 grams. Woodyatt (46) gives the average at 1 to 1.6 grams. These values for the human diabetic are considerably lower than those found for depancreatized dogs fed on sugar. Is it possible that they might be less in the latter, if the animals were fed exclusively on a meat diet?¹

This raises the question as to whether the glucose derived from protein is metabolized with equal facility to that derived from carbohydrate sources. Wilder, Boothby, Barborka, Kitchen and Adams (45) have noted that the same dose of insulin, given to a diabetic pa-

¹ Further discussion of the glucose equivalence of insulin on diabetic patients will be found in the clinical part of this review (page 249).

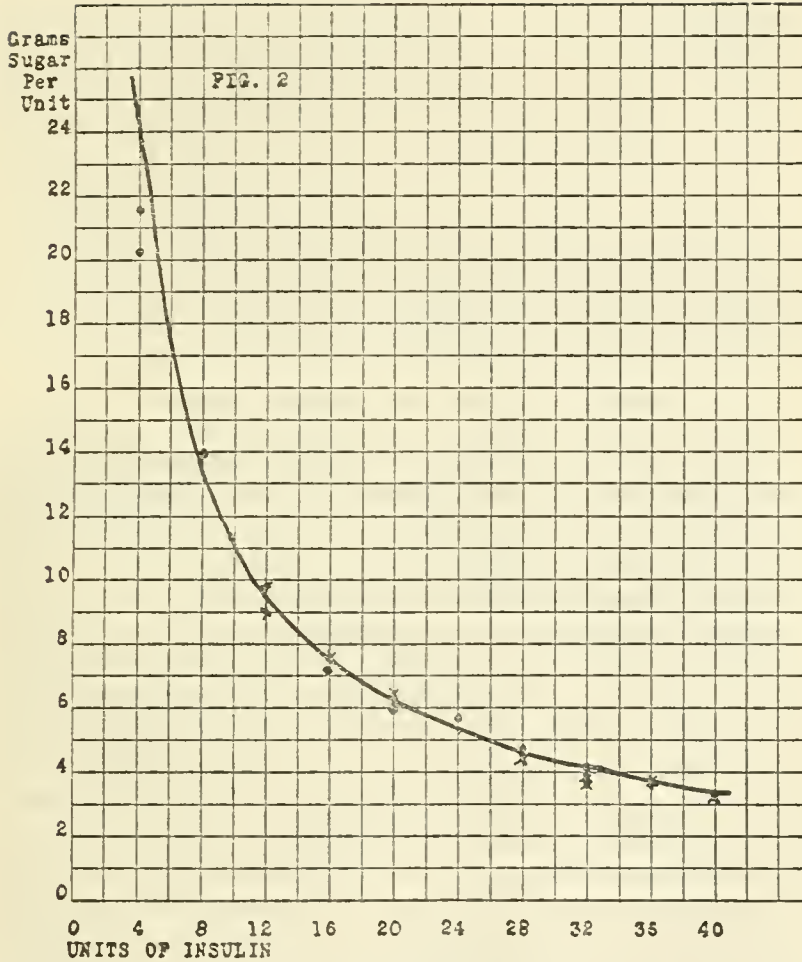


FIG. 2. CURVE TO SHOW RELATIONSHIP BETWEEN GLUCOSE EQUIVALENTS AND VARYING DOSES OF INSULIN

The curve corresponds to the equation $g(u)^{0.65} = 101.86$ and the dots and crosses give the results obtained in observations on the different animals.

tient at one period when the diet contained a high proportion of protein, caused much less reduction in the glycosuria than it did at another, when the protein was largely replaced by carbohydrate, the caloric

and total glucose values of the two diets being kept the same. Thus, while on a diet made up of 45 grams protein, 142 grams fat and 11 grams carbohydrate (i.e., 150 gram available glucose and 1956 cal.) 15 units of insulin reduced the daily sugar excretion from 57.78 to 9.46, giving a glucose equivalent of 3.32, whereas while on a diet of 159 grams protein, 118 grams fat and 41 grams carbohydrate (i.e., 145 grams available glucose and 1907 calories) 15 units only reduced it from 76.10 to 47.15, giving a glucose equivalent of 1.9. This would seem to indicate that the sugar derived in the body from protein is not so readily metabolized under the influence of insulin as sugar that is absorbed as such from the intestine.

EFFECT OF INSULIN ON NORMAL ANIMALS

It would serve no useful purpose here to enter into details with regard to the effects of insulin on normal and hyperglycemic animals, but a brief outline of the main results is necessary to an understanding of the highly practical question of its physiological assay. A full discussion of this subject will be found elsewhere (47).

Within a few minutes of the intravenous or subcutaneous injection of insulin into rabbits, the percentage of blood sugar begins to fall and it continues to do so at a practically uniform velocity for about forty- to sixty minutes. This "initial fall" is not noticeably influenced by the nutritional state of the animal until a certain low level is reached (about 0.045 per cent). Beyond this stage the curve either becomes less and less steep, or it begins to rise again as in well-fed animals, or it continues to fall, usually at a decreasing velocity, in animals that have been starved. The extent to which the blood sugar ultimately declines in starved animals depends partly on the dose of insulin, and under certain conditions the sugar may reach the vanishing point. These differences in the behavior of the curve are primarily dependent upon the amount of glycogen stored in the body, particularly in the liver. When the blood sugar has declined to a certain level, the glycogenolytic function of this organ becomes stimulated, so that more glucose is discharged into the systemic blood to replace that which has been lost. This stimulation occurs through the nerv-

ous system, since Burn (48) has found that when ergotamine is given along with insulin to well-fed animals the blood sugar curve fails to recover and behaves like that of a starved animal. This alkaloid acts like ergotoxine in paralyzing the endings of the sympathetic (thoracic-autonomic) nerve supply. We must suppose, therefore, that the so-called glycogenic center becomes stimulated when the blood sugar reaches a certain low level.

As to the cause for the initial fall in blood sugar, little is known for certain at present. As judged by the rate at which sugar disappears from blood after its removal from the body (glycolysis) insulin does not have any influence, either when it is added to the blood directly or when it is injected into the animal prior to bleeding (Eadie and Macleod (49)). We must conclude that the sugar disappears from the blood because of more rapid diffusion into the tissues, consequent upon a lowering of its intracellular tension. In other words, insulin must cause a sort of vacuum for sugar to be set up in the tissues, to fill which sugar is removed first of all from the blood and later, through the blood, from the glycogen stores of the liver. Further evidence that insulin accelerates the rate at which sugar passes into the tissues is afforded by the fact that its addition to Locke's solution perfused through the excised mammalian heart markedly increases the rate at which glucose disappears from the solution (Hepburn and Latchford (50)).

The question therefore, is: What causes the vacuum for sugar in the tissues? Since, as we have seen, insulin leads both to increased formation of glycogen and to increased oxidation of glucose when it is given to diabetic animals, it was natural to assume that the same two processes must be responsible for its disappearance in normal animals. With regard to glycogen formation, however, the evidence is very clear that insulin, instead of stimulating the process, has the opposite effect. Less glycogen is deposited, both in the muscles and the liver, when insulin is given along with sugar to previously starved animals than when the same amounts of sugar are given alone (McCormick and Macleod, (51)) and the glycogen in both these places becomes markedly reduced in amount when insulin is given to previously well-fed animals (Dudley and Marrian (52)). Regarding the possi-

bility that carbohydrate combustion is stimulated by insulin the evidence is not so clear, since observations on the respiratory exchange in different types of animal do not give entirely similar results. In dogs insulin causes a slight increase in R.Q. occurring as a rule during the first hour or so following its injection (i.e., during the time the blood sugar curve is falling rapidly) but this is followed later by a return to the original level. A rise occurs in the intake of oxygen, starting as a rule in about two hours after the injection of insulin, and the extent of this rise is more or less proportional to the dose. The volume of air breathed, the rate of breathing and the pulse rate all increase parallel with that of oxygen consumption and it can be noted that hyperexcitability of the animal and increased muscular tone, accompanied often with perceptible twitching, become evident at the same time and run parallel with the respiratory changes. These symptoms, as we shall see later, are related to the hypoglycemia and can be antidoted by giving glucose. When this is done the intake of oxygen does not become increased. In one dog, and to a less extent in another one, Pember and Dickson several times observed that R.Q. rose to over unity (1.2 to 1.3) following insulin, but as a rule, as already stated, this did not occur and at present we cannot account for the irregularity in the results, although there is reason to believe that it is related to the nutritional condition of the animal. In rabbits, following insulin, R.Q. also becomes increased and the intake of oxygen may either rise, or remain practically constant, or decline somewhat depending upon whether or not the hypoglycemic symptoms become prominent (53). In curarized rabbits, in which no muscular hyper-tonicity can develop, the O_2 -intake, according to Krogh (54) usually declines slightly. In mice there is a marked decline in the intake of oxygen (Dudley, Laidlaw, Treven and Boock (55)) and CO_2 , so that while R.Q. may rise this cannot indicate increased combustion of carbohydrate. In man, Kellaway and Hughes (56) and also Lyman, Nicholls and McCann (57) found that when insulin is given in sufficient dosage to induce the earliest symptoms of hypoglycemia, R.Q. becomes raised. According to Kellaway and Hughes the oxygen intake does not increase sufficiently so that the sugar which has dis-

appeared from the blood can be entirely accounted for by increased combustion of carbohydrate.

Taking these results as a whole the following conclusions concerning the mechanism of the action of insulin on normal animals seem warranted: (1) the amount of free glucose in the tissues (tension of glucose) promptly becomes reduced so that sugar is removed from the blood; (2) this reduction is not dependent upon a polymerization of glucose into glycogen; (3) it is not entirely due to an increase in the relative amount of carbohydrate undergoing combustion, although this may occur to a certain extent under, as yet undetermined, conditions.

It would appear, therefore, that insulin causes the glucose to be converted into some substance, that is hitherto unrecognized, into some substance which is not precipitated as glycogen but which at the same time has lost its reducing properties. Since it has been found that the inorganic phosphates of the blood become reduced at the same time as the reduction in blood sugar, it is possible that some substance related to hexose phosphate is formed. It has also been suggested that the high respiratory quotients sometimes observed after insulin indicate that fat-like substances are produced (cf. Macleod: Proceedings of the Institute of Medicine of Chicago, February, 1923). In this connection it should be noted that Dudley and Marrian did not find the amount of fatty acid in the liver to be changed in normal mice following insulin.

Hypoglycemic symptoms

When the blood sugar reaches a level of about 0.045 per cent, in rabbits, peculiar symptoms supervene. Preceded usually by a period of hyperexcitability, these then take the form of violent convulsive seizures, not unlike those of acute asphyxia, and between the seizures the animal may either remain in a hyperexcitable condition or gradually pass into one of coma which becomes more and more pronounced until death of the animal. In small animals, such as mice, coma and paralysis are the prominent symptoms. The symptoms in man are described elsewhere in this article (page 269). These symptoms are immediately removed by restoring the lost glucose to the blood. This

may be done either by injecting glucose subcutaneously, or intravenously, or by oral administration of any sugar which will yield glucose during its digestion. The latter method can of course only be adopted when the animal is still conscious and can swallow. Injection of epinephrin or pituitrin also removes the symptoms, the mechanism in the case of the former being that glycogenolysis is stimulated in the liver, thus adding glucose, endogenously, to the blood. This method should only be used on diabetic patients when it is certain that there is glycogen stored in the liver. Glucose is remarkably specific in its antidoting action. Of the other monosaccharides mannose and levulose are also effective and, to a less extent, galactose. The only disaccharide having any beneficial action is maltose, but this is somewhat delayed (Noble and Macleod.)

EFFECT OF INSULIN IN EXPERIMENTAL HYPERGLYCEMIA

As is well known there are in general two methods by which, apart from pancreatectomy, the percentage of sugar in the blood may be caused to increase. These may be classified as *exogenous* and *endogenous*. Exogenous hyperglycemia is brought about by injection of glucose subcutaneously or intravenously, or by feeding with any carbohydrate which is converted into glucose during the digestive process, and endogenous, by the various experimental procedures which are known to stimulate increased hepatic glycogenolysis. These procedures may be further divided into three groups, nervous, asphyxial and toxic. Among the nervous forms are puncture of the medulla oblongata, stimulation of the sympathetic nerve supply of the liver and the injection of epinephrin. The asphyxial hyperglycemias include those due to mechanical asphyxia, to poisoning by carbon monoxide and, to a certain extent, to the action of anesthetics. As examples of the toxic forms may be mentioned morphine and the anesthetics.

It is significant that insulin in suitable dosage is capable of preventing the development of hyperglycemia in all of these conditions (58). This fact is of importance in relation to the problem of the assay of insulin, which will be discussed immediately, and it is also of interest from the physiological standpoint. In the latter connection let us

briefly consider the antagonistic action of epinephrin and insulin. There are two mechanisms by which insulin might prevent the development of epinephrin hyperglycemia, these are: (1) by suppressing the hyperglycogenolysis; (2) by removing from the blood the extra sugar resulting from this process, as quickly as it is formed. In collaboration with E. C. Noble and M. K. O'Brien we have found, by measurement of the amount of glycogen in the liver in well-fed rabbits, that more of this substance disappears in a given time with epinephrin alone than with (the same amount of) epinephrin *plus* insulin, indicating clearly that insulin inhibits the hyperglycogenolytic effect of epinephrin. This result is of especial significance since, as we have seen, insulin given alone to well-fed animals itself causes hyperglycogenolysis. At the same time as it diminishes hyperglycogenolysis, however, insulin also accelerates the rate at which the sugar is removed from the blood, as is clearly shown by its effect on the exogenous form of hyperglycemia (page 232). These observations are quoted here to indicate the futility of attempting to explain the mechanism of the action of insulin until much more is known of its effects, both on normal and diabetic animals. There are already in the literature so many different views with regard to this question that it would occupy several pages to expound them and at the end we should be no nearer the truth.

PHYSIOLOGICAL ASSAY OF INSULIN

A unit of insulin, as originally defined, is that amount which can lower the blood sugar of a normal rabbit weighing 2 kgm. to the convulsive level within four hours. More recently the definition has been modified to the extent that the animals must be deprived of food for twenty-four hours prior to being used—so as to reduce the glycogen stores to an approximately uniform low level. Since it was stated by some clinical observers that there are certain types of cases of diabetes in which less than one (physiological) unit of insulin as above defined is sufficient, it was decided, on their recommendation, to consider as one (clinical) unit for practical purposes one that was one-third of the above strength.² Even when precautions are taken to use animals

² In this article, unless otherwise stated, one unit means the physiological and not the clinical unit.

of uniform size and breed, and to render them free of glycogen by more prolonged starvation than one day, the assays are, however, not strictly accurate. To minimize the error it is therefore necessary to use large numbers of animals and to base the final assay on the average of the results. In order to avoid the necessity of having to make the numerous blood-sugar estimations which this would entail, some observers have based their assays on the incidence of the well-characterized symptoms, on the assumption that these correspond to a definite lowering of blood sugar. This method has been used on rabbits in the Eli Lilly Laboratories, varying quantities of a given preparation of insulin being injected into a number of animals and the unitage calculated from the proportion which developed symptoms within a certain time. The method, however, is a costly one, and since, if the development of symptoms is to form the sole basis for assay, it is evident that smaller animals than rabbits might be used, attention has been paid by D. R. Fraser (59), A. Krogh, and H. H. Dale, and J. H. Burn (60) to the possibility of using mice. It has been found that when these animals are injected with insulin and kept at room temperature the most constant results are a profound fall in body temperature and weakness, so that the animal sprawls out on its belly, convulsive symptoms being only occasionally observed. If the fall of temperature be prevented by placing the animals in an incubator at 28°C., however, the symptoms are much more like those described for rabbits—convulsions, coma and paralysis of the legs being evident. The symptoms are immediately removed by injecting glucose. By taking precautions to standardize the diet which is given prior to a short period of starvation (eight hours) and to use animals of similar age (varying from 16 to 20 grams in weight) a method of assay has been evolved in which one mouse unit is considered as the amount of insulin which will cause definite symptoms in 50 per cent of the injected animals (Krogh). At present certain difficulties make this method of assay unsatisfactory, but it is hoped that these may soon be overcome, since it is obvious that it would be of great value to be able to use these animals in place of rabbits.

There are, however, several serious objections to the use of any method of assay that is based on the incidence of convulsions. The chief of these are: (1) that different animals, although of the same

size, sex and breed and in the same nutritional condition are not necessarily of equal susceptibility to the development of symptoms, following the same dose of insulin. (2) that no consideration is taken of the duration of the hypoglycemic effect.

With regard to the first of these objections it should be pointed out that the symptoms make their appearance when the blood sugar has reached about 0.045 per cent, as originally stated by us, only when nonstarved rabbits are used. With starved animals, the blood sugar is usually considerably below 0.045 per cent when symptoms supervene (cf. McCormick, Macleod, O'Brien and Noble (61)). Thus, out of a total of 335 rabbits injected with insulin no symptoms were noted in 64 in which one or more of the blood sugars was decidedly below 0.040 per cent. On the other hand it is very seldom indeed that any symptoms are observed in starved animals before the blood sugar has reached 0.045 per cent (Macleod and Orr (62)). The relationship between symptoms and blood sugar may therefore be correctly stated to the effect that they are likely to occur when the percentage of the latter has fallen to 0.045. In the light of more recent work it is probable that the frequency of symptoms at the higher levels of blood sugar, as observed by us in the earlier work, was due to the fact that the animals were not starved and therefore contained considerable stores of glycogen. By mobilization of glucose from these reserves, when the hypoglycemia reaches a certain critical level (see page 222), a further tendency to the development of hypoglycemia would be partially counteracted and the blood sugar thus held at a level (of about 0.045 per cent) at which toxic effects only slowly supervene. In starved animals, on the other hand, the blood sugar continues its rapid descent and goes below the level at which symptoms *may* appear before sufficient time has elapsed for the toxic condition to become developed.

The objection that no estimate is given of the duration of the hypoglycemic effect, when the assay is based on the incidence of symptoms, is of importance. As has been pointed out elsewhere in this article this duration depends very largely on the amount of glycogen stored in the body. Even if we could be certain that the glycogen had been reduced, by starvation, to the same extent in all the observed animals it would not necessarily follow that with different

preparations of insulin the hypoglycemia must always be of the same duration. It is possible not only that the rate of absorption of insulin may vary, but also that its hypoglycemic action may be antagonized or delayed by the presence of extractives from the pancreas which have a hyperglycemic effect. In this connection it is of importance to remember that we have observed that extracts of the zymogenous pancreatic tissue of certain of the bony fishes causes an increase in blood sugar, islets being practically absent from this tissue but existing as separate glands (principal islets of Diamare and Rennie) in the mesentery (Macleod (29)). Collip (63) has also observed that insulin prepared by the usual methods from yeast, clams, etc., may not cause a marked degree of hypoglycemia until many hours after injection. It has also been our impression that impure preparations of insulin have a more prolonged hypoglycemic effect than those that are highly purified. For all these reasons, therefore, it is evident that the incidence of convulsions is not likely to prove a suitable method upon which to base the accurate assay of insulin.

Before proceeding to discuss the method dependent upon the behavior of the blood sugar it may be well to point out further that one might have a preparation of insulin which contained, in 1 cc., an amount of insulin that was sufficient to cause a considerable reduction in blood sugar and yet not bring this to the convulsive level. Although such a relatively weak preparation might conceivably be assayed by the convulsive method, as by giving large injections, it is evident that the hypoglycemic effect could be arrived at much more satisfactorily by observing the behavior of the blood sugar.

The method of assay used at present by the Insulin Committee of the University of Toronto is carried out as follows: Normal blood is removed from the ear vein of each of 9 rabbits which have been starved for twenty-four hours, and the percentage of blood sugar is determined. No rabbit is used for the assay whose normal blood sugar is found to be below 0.105 or above 0.125 per cent. Into 1 of the rabbits is injected, subcutaneously, an amount of insulin which is expected to correspond to one physiological, or three clinical units. Into a second animal is injected an amount, about 50 per cent greater. The other 7 rabbits are injected with amounts varying from slightly below one unit to slightly below one-half unit. At intervals of

one and a half, three, and five hours after injection about 1 cc. of blood is again removed from each of the rabbits and the blood sugar determined. The results are then calculated by the use of a formula which takes into consideration all features of the definition of a clinical unit, as well as allowing for the duration of the effect and for varying weights of the animals. This formula is:

$$\text{Clinical units per cubic centimeter} = \frac{a}{b} \times \frac{w}{c} \times 1.5$$

where a is the difference between the percentage of normal blood sugar and the average of the percentages of the blood sugars at one and a half three and five hours after injection; b is the difference between the percentage of normal blood sugar and 0.045; c is the number of cubic centimeters of insulin injected, and w is the weight of the rabbit in kilograms. The factor 1.5 is used because the original (physiological) unit was based on the effect produced in a rabbit weighing 2 kgm. and the clinical unit is one-third of this (i.e., $\frac{1}{3}$). In several of its details the formula is more or less arbitrary. This is the case mainly with regard to the value of a , the other values b , c and w being required merely so that the assay may conform with the definition of a unit. Thus, the value a is taken as a measure of the average extent to which the blood sugar becomes lowered over a period of five hours following the injection of insulin. It might therefore be the same in two cases in one of which the blood sugar had quickly fallen to a low level with prompt recovery and in another in which the initial fall was not so steep but the recovery more prolonged. These differences are dependent, as has already been explained, partly on the purity of the insulin preparation, partly on its rate of absorption and partly on the fact that the mobilization of glucose from the glycogen stores of the liver is not necessarily the same in different animals, even when these have been starved for twenty-four hours. Probably the most important reason for this is that the liver in some of the animals will be devoid of glycogen whereas in others it will contain a small but varying amount. It is possible that starvation for a longer period preceding the injection—several days for example—might minimize this source of error but since this weakens the animals it would not be a justifiable procedure unless it decidedly minimized the errors and this, in our experi-

ence, is not the case. Even supposing the glycogen were equal in amount in different animals, another possible source of inconstancy in the rate of recovery of blood sugar is varying sensitivity of the controlling mechanism by which this recovery is brought about. When there is only moderate decrease in blood sugar it is conceivable that the threshold level (of hypoglycemia) necessary to excite a marked increase in glycogenolysis is not reached, the recovery in blood sugar which does occur in such a case being dependent on the normal sugar production from the liver. On the other hand when the initial hypoglycemia occurs rapidly and is pronounced, hyperglycogenolysis becomes violently excited with the result that much glucose is added to the blood and recovery of blood sugar is therefore prompt.

Quite apart from these physiological errors there are others dependent on the mathematical inaccuracy involved in taking the average of the blood sugars at one and a half, three and five hours. What we really desire to know is the extent of the area in the curves during which the blood sugar is below the normal level and it is clear that we do not accurately measure this by the above method. On the other hand, however, with such dissimilarity as exists in the results obtained on different animals towards insulin it is difficult to apply any other method of evaluation. The choice of the intervals for the first analysis of the blood sugar, following insulin, was based on the observation, along with G. S. Eadie (64), that the maximal influence of insulin in combating the development of hyperglycemia due to the injection of glucose (2 grams per kilogram body weight) is most marked when the glucose is injected in about one and a half hours following the insulin. The interval of five hours was chosen for the last analysis because it was found that by this time the effect of insulin, on starved animals, has usually disappeared, the three hour interval being chosen as intermediate between the two others. In the light of later experience it would probably be more accurate to make the first analysis in one hour after the injection since at this period the blood sugar in the majority of starved animals is at or near its lowest level. This modification is now being introduced but will not be formally adopted in the assays of the Insulin Committee unless it is found that by so doing a greater degree of accuracy is attained.

Another source of inaccuracy in the present method is dependent on the fact that it is assumed that the extent to which the blood sugar becomes lowered is directly proportional to the dose of insulin or, in other words, that the fall in blood sugar will be twice as great when the dose is doubled. This is certainly not the case since the action of insulin on the blood sugar, like that of epinephrin on the blood pressure (cf. Murray Lyon) obeys a logarithmic law, the effect of a unit becoming progressively less and less as the dosage is increased. It will be recalled that Frank N. Allan has shown a similar relationship to exist for the glucose equivalents of insulin on depancreatized dogs (page 218). We have endeavored to obtain adequate data from which to correct the present method used for the evaluation of a but without sufficient success to warrant the change. The reason for these difficulties will be perfectly evident when the blood sugar curves of a number of starved rabbits injected with the same or with different doses of insulin are examined. Even when the same dose of the same insulin is injected, at weekly intervals, into the same rabbit the degree of hypoglycemia which results may not be the same (Macleod and Orr) so that it is hopeless to expect that varying doses will give dependable results. There are evidently certain factors involved in the reaction of an animal to insulin which we do not as yet understand and cannot therefore control. One of these may be related to the body temperature. It is significant that A. Krogh has found that in mice temperature greatly affects the susceptibility towards insulin (page 228) and we have noted that in rabbits the hypoglycemic condition supervenes more rapidly when the animals are kept warm. Even when care is taken to keep the rabbits under similar conditions with regard to the cooling condition of their environment, however, the effects of insulin are not uniform. It is possible that more uniform results will be obtained by using animals of the same breed, age and sex subjected to similar experimental conditions. We are at present investigating this possibility with standard white rats of the same litter.

It is realized, therefore, that the above factors make the evaluation of a a matter of considerable uncertainty so that means must be taken to minimize the error thus entailed in the final assay. This is done by employing a sufficient number of animals, and at present the practice is that if five out of the nine assays do not vary by more than 25 per

cent an average of these five is taken as the final assay. Needless to say, if five out of nine assays do not correspond within these limits the assay is repeated until a majority do so. The error is then probably not more than 10 per cent between extremes. As a general rule the higher values are obtained when small amounts of insulin are used (c in formula) which of course indicates that the relationship between dosage and hypoglycemic effect is a logarithmic one (see page 220). Sometimes with the smaller doses a result is obtained that is very much higher than the average of the others of the same group. In such cases the assay is repeated on three more rabbits using this dose, and unless the result is duplicated the exceptional assay is disregarded. The present tendency in carrying out the assays is to use a limited supply of rabbits, a record being kept of the exact behavior of each animal in previous assays. By so doing we are really making the assay by comparing the effect of an unknown sample of insulin against that of one whose effect on the same animal is already known. Although this probably gives more constant results these are not entirely satisfactory, for reasons which have already been discussed. These so-called "registered" animals are used for assay purposes once a week and it is of interest that they gain rapidly in weight and after some time begin to develop a decided resistance towards insulin, the blood sugar being lowered to a much less degree than previously was the case with the same dose, per kilogram body weight, of the same insulin. The occurrence of convulsions does not appear to interfere in the slightest degree with the well-being of the animals but nevertheless it is the practice to avoid, as much as possible, giving such doses as will cause them. The only way by which reliability of the assays can be tested is by comparing those made on different occasions from the same batch of insulin.

Comparison of the physiological assay with the clinical action of insulin

At first sight it might appear an easy matter to determine the number of grams of glucose which each unit of insulin could cause to be utilized by a diabetic patient, the so-called glucose equivalent. For reasons set forth elsewhere in this article, however this cannot be

done unless the total dosage of insulin and the carbohydrate balance of the patient are taken into consideration and even then it is likely to be inconstant because of the varying endogenous production of insulin from what still remains of the patient's islet tissue. In a very general way it is usually considered that in cases of diabetes of moderate severity one clinical unit of insulin is capable of causing about 1.5 grams of glucose to be metabolized. Even at the risk of repetition it should be pointed out, however, that many more grams of sugar would be metabolized per unit when one unit of insulin is given to a patient excreting large quantities of sugar than when several units are given. With each added unit less and less glucose *per unit* would disappear from the urine. For these reasons, therefore, close correspondence between the physiological assay and the clinical evaluation of the glucose equivalence of insulin, as this is ordinarily determined on diabetic patients, cannot be looked for. On depancreatized dogs, on the other hand, the two assays run closely parallel partly because there is no possibility of the endogenous production of insulin and partly because the dietetic conditions can be adequately controlled.

ULTIMATE FATE OF DEPANCREATIZED DOGS TREATED BY INSULIN

The clinical history of the depancreatized dogs that were used for determination of the glucose equivalents (page 220) has revealed several facts of interest. As has already been stated these animals remained in excellent nutritive condition and usually gained somewhat in weight while on a diet of an excess of lean meat (500 to 600 grams daily) and cane sugar (100 to 150 grams daily). The only unphysiological condition observed, apart from glycosuria and hyperglycemia, was that only about one-half of the ingested protein was absorbed, as judged by the excretion of nitrogen in the urine. Of 3 such animals, 1 lived for two months, a second for four, and the third for seven months following the pancreatectomy. After these periods, in each case, symptoms of an acute breakdown in the hepatic function supervened—bile in the urine, jaundice, refusal to take food—and great bodily weakness soon developed, ending in the death of the animals within a few days of the onset of the symptoms. In all cases the liver was found at autopsy to be invaded by large quanti-

ties of fat—over 35 per cent of the moist weight being found in one case. On microscopic examination the fat was found to exist as very large globules, particularly in the cells of the portal area, giving the sections an appearance not unlike that of adipose tissue. In the few cells that contained relatively less fat, near the central vein, the protoplasm and nuclei showed some evidence of degeneration. The symptoms and the pathological findings strongly suggest the action of some highly toxic agency on the liver, but it is interesting that the kidneys of the 2 animals that lived four and seven months, respectively, showed no signs of toxic changes, although such was observed in those of the animal which died in two months.

There are several possible causes for the toxic condition. It may be related to the faulty absorption of food, due to absence from the intestine of the pancreatic juice. That protein was most imperfectly absorbed has already been stated and it is possible, as has also been suggested by Dr. Almon Fletcher, that the bacterial digestion of the unabsorbed protein would cause toxic decomposition products to be absorbed into the portal blood and so act injuriously on the liver cells. Lombroso (64a) has also noted that the absorption, particularly of fat, becomes very imperfect in depancreatized dogs although it is little impaired provided some pancreatic tissue—a subcutaneous graft, for example,—be left in the body, without any pancreatic juice gaining the intestine. McClure, Vincent and Pratt (64b) have denied this influence of pancreatic tissue on the efficiency of absorption of fat and have stated that this remains tolerably normal in the entire absence of pancreatic tissue from the body. We did not determine the extent of fat absorption in the above mentioned animals but we are certain that it was greatly impaired, it being necessary in order to keep them in good condition to remove as much fat as possible from the meat which was fed. It is possible, therefore, that owing to the entire absence of pancreatic tissue from the body there comes to be a deficiency of lipase and that this leads ultimately to a break-down in the process of fat metabolism and accounts also for the faulty absorption of fat from the intestine which would otherwise proceed with tolerable efficiency, because some of the lipase carried by the blood would find its way through some of the secretions into the intestinal tract. Finally,

however, it must be remembered that jaundice in dogs, if not due to obstruction of the larger bile ducts, or to blood changes, is believed to be invariably associated with degeneration of the hepatic cells which may be due, in these experiments, to toxic substances derived from the bacterial decomposition of protein in the intestine.

PART II. CLINICAL SECTION

W. R. CAMPBELL

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INTRODUCTION

The experiments of Banting and Best in Toronto during the summer of 1921 were regarded with much interest by those conversant with them. The remarkable results of the crude extracts, (19, 22) the product of their experiments, upon depancreatized animals early encouraged the hope that in due time the essential deficiencies produced by ablation of the pancreas in animals might be controlled, thus contributing vastly to our knowledge of the physiology of carbohydrate metabolism and furnishing a basis for a renewed attempt at treating human diabetics. How abundantly that hope has been fulfilled it is perhaps unnecessary to relate here. We can thankfully echo Joslin's words in the introduction to *diabetes* in Oxford Medicine—"insulin is here!" May we also hope—to stay, or perhaps one day to be succeeded by one of its isomers or derivatives, robbed of certain of its limitations and with certain of its therapeutic properties enhanced.

For human physiology the crucial experiment must always be on man himself and in one sense the physician ranks as a physiologist of greater or less degree. But not alone is the physician desirous of knowing. His prime duty is to relieve, mayhap to cure, and experimentation on man is not justifiable without substantial hope of ultimate reward in knowledge or information conducive to that end. The splendid collaboration of the group of workers in the Department of Physiology swept the animal researches rapidly forward. When, by means of animal experiments, most of the essential points had been demonstrated, the Department of Medicine was requested in November, 1921, to test the pancreatic extracts in relieving human disease.

The supply of degenerated glands of dogs after pancreatic duct ligation and of fetal calf glands was quite insufficient to provide an extensive trial in man of the extracts. For this purpose it became necessary to use extracts of adult beef pancreas, the uncertainty of the supply of which, coupled with the pressure of other physiological problems, delayed the actual clinical trial of the extracts until January 11, 1922. The first person to whom this latter extract was administered was a boy of fourteen years referred to the writer by

Dr. Skeeles of Toronto. For a month prior to the actual administration the patient had been kept on a strict starvation diet. The records show that, while fasting, the blood sugar varied between 0.5 and 0.6 per cent. Sugar was continuously present in the urine and, indeed, the blood sugar level throughout the day scarcely varied. The patient exhibited ketosis to a marked degree. On January 11, after blood had been taken for sugar estimation, the pancreatic extract—a murky, light-brown liquid containing much sediment, which dissolved to a considerable extent on being warmed—was injected into the buttocks. The amount administered was 15 cc., a dosage calculated as being one-half as much as would give a definite reduction in blood sugar in a depancreatized dog of equal weight. Somewhat to our disappointment the blood sugar level was reduced by only 25 per cent. Looking back one can see as well a slight fall in sugar excretion, but at the time this was too small to be convincing.

Production of sterile abscesses in the first patients to whom the extract was administered made it imperative that the mixture should be rendered protein free. The desired result was accomplished by Collip who, by precipitation of the insulin, was also able to supply stronger extracts not containing the large amounts of salt and protein which in the earlier extracts had caused the patients to complain of a persistent burning sensation. The chemical conditions essential for production of this extract were not well understood and at this time none of the patients received nearly adequate treatment.

Various points were investigated in different patients and sufficient data obtained to justify our preliminary statements (30, 65) regarding the effect produced on some of the fundamental phenomena of diabetes mellitus by treatment with these extracts, namely: that blood sugar can be reduced to normal values; glycosuria can be abolished; the acetone bodies can be made to disappear; the respiratory quotient can be made to rise decidedly and the patients themselves to remark their improvement. At this date, moreover, the comparatively short duration of the action of the extract, the occurrence of toxic reactions and the limitation of its value to certain types of diabetes were referred to.

Meanwhile, through the work of a team of workers (23, 24, 66, 35, 32, 67) in the departments of Physiology and Pathological Chem-

istry, the problems of standardization of insulin and its effects on laboratory animals were prosecuted diligently. As a result it was found that the lowering of the blood sugar, which was first observed by Collip to occur in normal rabbits, could be made the basis of assay. It was also found that the peculiar toxic symptoms which supervened after large doses of insulin were dependent upon the hypoglycemia and could be immediately antidoted by glucose. Coördinated rapid solution of particular problems, coupled with prompt communication of the results to all members of the group, encouraged all to believe that the individual difficulty confronting each worker would soon be surmounted and to hope for the ultimate production of sufficient extract to undertake more detailed investigation of the problem in man.

Shortly after the publication of the preliminary clinical report an attempt was made to produce insulin on a larger scale. Unfortunately, though small quantities could be obtained by reverting to the experimental laboratory apparatus, a great many failures resulted before the larger sized apparatus gave practical yields, and the clinical work by the Department of Medicine was virtually discontinued for a time.

During the summer of 1922 investigation of insulin was started in the Christie Street Military Hospital and in the Hospital for Sick Children, Toronto, by Dr. F. G. Banting, and recommenced in the Department of Medicine of the Toronto General Hospital. As soon as it became possible to obtain pancreatic extract, or insulin as it was now called, for additional patients the most severe cases were admitted and treated. Those who through circumstances were unable to pay, received treatment in the public wards of the Toronto General Hospital, while an equal number of the more affluent were treated in wards especially reserved for them. By this means it became possible to limit our attention to a series of most severe diabetics who not only needed the insulin the most but also furnished the most searching test of its efficiency. The patients all realized that they were embarking on an experiment which might be of great benefit not only to themselves but to other sufferers from the disease and that a most strict adherence to the minutiae of treatment was necessary if the results were to be of any value. To

their credit it may be said that I have not previously experienced such satisfactory coöperation on the part of patients. Thanks are due to the authorities of the Toronto General Hospital, who provided special nurses, extra dietitians, assistants and supplies sufficient to carry out this investigation and coöperated in every possible way to make it a success.

We were certain that, if insulin were properly applied, a new chapter in the treatment of diabetes would be opened. How to prevent it from being exploited and, through ill-advised treatment, from falling into disrepute was a problem which the Insulin Committee solved by placing it in the hands of some of the eminent specialists on diabetes in the United States. Owing to the supplies being limited at first it was necessary to restrict its distribution to a few, but later the numbers investigating its use were gradually expanded as the production of insulin increased. To these, our collaborators, we are grateful for their kindly assistance and loyal support. Among so many with somewhat differing views regarding diabetes, there have naturally arisen differences of opinion as to methods of treatment, but underlying all these is a fundamental confirmation of our original thesis (30, 65), which has firmly established the therapeutic value of the drug.

The second clinical paper (68) on insulin was based on experience derived from the treatment of more than 50 patients with severe diabetes. In general it may be said that the results of the treatment of the larger number of cases completely confirmed the tentative conclusions reached in the preliminary paper. Adequate supplies of insulin permitted a much more prolonged trial in a number of individuals and showed that no untoward effects need be anticipated from its continued use. Glycosuria was successfully controlled and the blood sugar maintained at normal levels for long periods. The acetone bodies were burned under the influence of the insulin and many cases of severe acidosis recovered. Six patients in whom complete diabetic coma had developed recovered after its use, while four died, one due to insulin shortage and the other three to complications. Uncertainty of standardization, deterioration of extracts, as well as imperfect balance between insulin and ingested carbohydrate gave rise to a considerable number of peculiar reac-

tions associated with hypoglycemia in patients under insulin treatment. This condition in man, which in many ways resembles the hypoglycemic state in animals, was studied and described together with its treatment.

Notwithstanding the excellent control of the chemical phenomena of diabetes the greatest interest centers in the improvement in the general condition, mental and physical, of the patient. In these severe cases the improvement was remarkable and can only be compared to that produced by another internal secretion—thyroxin in myxedema cases. The mental state of hopeless, irritable depression, almost melancholia, characteristic of the advanced, emaciated diabetic gives way to a spirit of cheerfulness and optimism. Insomnia becomes less troublesome and interest in his surroundings is reawakened. The feeling of lassitude or early exhaustion after slight exercise disappears. The carriage of the patient improves and physical vigor is gradually restored. Thirst is no longer excessive and a state of continuous hunger is replaced by a sense of comforting fullness with an appetite for meals. The body weight can be caused to increase at will and many of the patients in a very short time have been able to return to their former work. In the case of patients with infections insulin was found to confer a definite benefit. A number of suggestions as to the use of insulin in treatment are incorporated in the paper.

In the two earlier papers a number of points received only brief mention. More detailed presentation of the results of the investigation of insulin appeared with the papers of our earlier collaborators in the November–December, 1922, number of the *Journal of Metabolic Research*. The remainder of this article will be concerned largely with a review of these papers (69, 34, 70, 71, 45, 72, 73, 74, 46, 75) since in them is to be found first reference to the effect of insulin on certain phenomena of diabetes. To many other authors we are indebted for confirmatory data which it is not possible to review in detail.

General information

Insulin, a substance widely distributed throughout the animal kingdom, is usually derived from the pancreas of mammals or fishes.

Its function is to promote the combustion of carbohydrates. It is obtainable commercially as a sterile standardized solution in various strengths, five, ten and twenty units per cubic centimeter. It also can be prepared in the form of hypodermic tablets (Krogh, McCormick and Noble (60)). Apart from the decreased cost it may be expected that these will prove of value because of their greater stability especially in very cold or very hot climates. Under ordinary conditions of temperature insulin solutions are stable for long periods without especial precautions. The physiological assay of insulin is discussed in the first part of this paper and hereinafter the clinical unit is the unit of measurement of the hormone.

Administration of insulin

Insulin, as was shown by Banting and Best in their original communications (19, 22), is easily destroyed by incubation with pancreatic juice. Peptic activity likewise rapidly destroys its action. Tests of insulin administered by mouth have been made by Banting, by Woodyatt (46) and by Joslin (71) with negative or almost negative results. The slight and variable activity found in some instances when given by this pathway is probably dependent upon early absorption of a portion of an enormous dose. Woodyatt (46) also conducted experiments in which oral, rectal, vaginal and intranasal administrations were given as well as inunctions. These methods yielded very weak, doubtful or negative results. My own experiments (unpublished) with insulin by inunction show a slight activity of the substance when administered in enormous doses. Telfer (76), in rabbits, obtained positive results after rubbing insulin into the shaved skin of the animal. These results have been duplicated by Macleod and McCormick (60). The procedure deserves further investigation, both as a means of prolonging the action of insulin and also as a method of producing a quite definite feeling of well-being in some patients who do not show distinct evidence of requiring insulin. For this purpose a much cheaper and less refined preparation could be used.

In the average case of diabetes insulin is best administered hypodermically. The intravenous method is only used to obtain rapid

effects in cases suffering from diabetic coma. The needle should be fine and very sharp— $\frac{7}{8}$ inches 27 to 29 gauge needles are the best—and the insulin injected through cleaned skin into the subcutaneous tissue. A little pressure may be used to numb the skin but great care should be taken to cause as little trauma as possible. Particular attention should be given to the introduction of the needle into loose subcutaneous tissue. Any part of the body where this is available is suitable; pressure points should be avoided. Insulin must not be injected into the skin as marked redness and stinging and sometimes, in the emaciated patient, necrosis of the skin develops. In all probability this is due to the preservative used. The injection must not be made into the muscle tissue as it is not only painful but gives rise to swelling and long continued induration. Bowie and Robinson (77) have shown that degeneration of the muscle fibres with a leucocytic invasion occurs in such areas. Similar microscopic appearances are presented by many other substances injected into muscle. The ultimate condition appears to be fibrosis in the muscle.

The time of administration of insulin is important in relation to the size of the dose and the efficiency with which it supplements the internal secretion of the patient's pancreas. Under normal conditions the blood sugar in man is maintained at relatively constant levels save for periods following meals, during which food is being absorbed. Coinciding with this we have, in all probability, a continuous production of insulin with a peak of production at the period of absorption of carbohydrate. In cases of mild diabetes these latter periods are the only ones in which the production is likely to prove insufficient so that glycosuria results. With severe diabetes glycosuria is constant, though the percentage excreted after meals is higher than at other periods of the twenty-four hours. In using insulin it would, of course, be ideal if it could be supplied so as to imitate the natural process but this would mean continuous injections or adoption of some method of delaying absorption. Experience has shown that insulin can develop its full therapeutic value when it is given at any time of the day provided a suitable amount is used for the particular needs of the individual. For the large majority of patients, however, insulin is best given in the half hour just pre-

vious to meals. This permits the insulin to neutralize the hyperglycemic effect of the meal. In most cases, with diets containing somewhat less than one-third of the carbohydrate in the breakfast, one-third in the noon meal and the remainder in the evening meal, it is possible to divide the insulin into two equal doses and administer these before breakfast and before the evening meal.

In certain patients, however, the proportion of the insulin required at each injection may not be the same, and it has to be remembered that hypoglycemia must be avoided just as carefully as hyperglycemia and glycosuria. The morning dose of insulin given when the blood sugar is at the fasting level is absorbed in sufficient time to prevent glycosuria as a result of breakfast, and the latter delays the fall of blood sugar due to the insulin. When minor hypoglycemic reactions do occur they are most often noted at 11:30 a.m. to 12:00 noon, and are quickly antidoted by any easily absorbable carbohydrates present in the noon meal. Lyon's (78) apparently different results are, I believe, dependent upon the fact that he permits high morning fasting blood sugars so that glycosuria occurs when the insulin is given at the same time as the meals. He recommends insulin two hours before meals. This is an inconvenience which should be avoided in the case of most patients.

The period from the evening meal to breakfast is the longest time elapsing without replenishment of the carbohydrate supply. Provided sufficient insulin is available when energy is required, carbohydrate is burned in preference to other foods. This process may use up the supply of carbohydrate and leave only fat and protein to be burned during the early morning hours. It is not unusual for a slight ketonuria to develop at this time. The same process might cause a greater liability to hypoglycemia during this period and this condition might be very dangerous if it occurred during sleep. For these reasons a larger portion of the carbohydrate is prescribed in the evening meal.

Should the insulin requirement exceed forty to fifty units it is usually better to divide the day's allowance into three or more doses. Unless these patients are receiving diets quite high in total glucose they will usually be quite severe cases with little or practically no tolerance for carbohydrate, and the most suitable time for injecting

the doses may require very careful investigation. Often they may be given just before each meal, though the quantities should not be equal at each meal.

The practical utility of a method in which but one dose of insulin is administered daily has led Fletcher and the writer on many occasions to test its efficiency. When large doses must be given this method is unsuitable since the patients develop hypoglycemia more often, and when the relatively short period of the action of insulin has passed, hyperglycemia, and often glycosuria, develops. With one dose, also, the total amount of carbohydrate burned is less than with several doses. The carbohydrate equivalent of small doses of insulin has also been shown by Dr. Frank Allan, (43) working in Professor Macleod's laboratory, to be decidedly greater than when larger doses are given. Dr. Frederick M. Allen's (75) objection may be quoted: "Even if all food in excess of the patient's tolerance be added to a single meal following this injection the fact remains that this food is only partially burned at the time and is largely stored as glycogen and other reserves which create demands for insulin after the single dose is used up." He favors two doses in the milder cases, three (before each meal) in the more severe, and four (i.e. every six hours) in the most severe cases. In connection with the last class of diabetic it should be noted that, in one most severe case apparently with zero tolerance, insulin was given as often as every four hours.

Other investigators have administered insulin somewhat differently. Fitz, Murphy and Grant (73) give it two to three times daily, an hour to an hour and a half before meals. Williams (72) has included charts in his article showing the superiority of three doses of insulin daily. Woodyatt (46) expresses the view that one injection daily is sufficient in 90 per cent of his cases, though occasionally two doses are employed. Wilder (45) believes that maximal results are obtained with a single injection, thirty minutes before breakfast. Barborka (79), writing later from the Mayo Clinic, favors a single dose when this does not exceed thirty units per day, and twenty-five units before breakfast with the remainder of the insulin at 3:00 p.m. if more than thirty units are necessary. Joslin (71) and his associates make use of two doses of insulin before breakfast and supper in the majority of their cases.

We have not been able to obtain satisfactory results with the single dose method when the daily requirement exceeds ten units. Objection has already been taken to some of the methods mentioned above on theoretical or practical grounds. These objections are obviously founded on ideals of treatment of the diabetic. Since there is not uniformity of opinion among the various groups of workers who have used insulin, with regard to methods of administration, it may be of service to note here the general principles which have guided the treatment as given in Toronto. In the opinion of the writer these include the principle of adequate nutrition and sufficient food on which to do light work since, failing cure, this alone accomplishes the maximum result—conversion of the patient into an economic asset. Adequate rest to the pancreas is essential to prevent deterioration of tolerance and can only be accomplished by keeping the patient aglycosuric and his blood sugar reasonably near the normal level. Excessive caloric intake in all diabetics should be discouraged. Maintenance of patients at or 10 per cent below the normal weight is desirable. For many cases insulin will be necessary to accomplish these results but for a very considerable proportion of patients dietetic treatment alone is necessary.

The fundamental principle underlying treatment is thus a therapeutic one and we are the less concerned over the question of under-nutrition since insulin can be used to replace the wanting endogenous secretion of the pancreas in those patients who previously could not be made aglycosuric without subjecting them to marked under-nutrition. That marked under-nutrition possesses any mysterious benefits for the patient who has sufficient tolerance, after eight years' use and observation of the method, I frankly do not believe. Why it should now be employed when insulin is available is not clear. Arguments based on the markedly increased amount of insulin required for maintenance of a patient aglycosuric when on higher diets are wholly fallacious since they take no account of the chemical law underlying the action of insulin. (See preceding pages (Frank Allen, pp. 218 et seq.))

Sensitization to insulin

In the earliest preparations sensitization to the proteins of the particular animal furnishing the pancreas was to be feared and, when our first patient returned to us in a state of severe acidosis bordering on coma with very marked dehydration, considerable care was taken to avoid producing an anaphylactic reaction. Desensitization precautions (Besredka method) were used. Fortunately no evidence of sensitization has appeared in this patient, though it is now two years since pancreatic extract was first given, and insulin has been given daily during the last sixteen months. In the large majority of patients sensitization phenomena have been entirely absent. The occasional case, however, has sometimes shown a slight urticaria and, indeed, one particular batch of insulin, produced in the hottest part of last summer, showed particularly irritant qualities for a number of patients. Much more rarely marked swelling, itching, wheals, or even pain has been produced. Local applications of saturated magnesium sulphate solution, saturated soda bicarbonate solution and 1 per cent phenol have given local relief in these cases. Adrenalin has been tried with temporary success. Calcium chloride internally has not proved of value. One's impression is that these phenomena are steadily diminishing in number with the increasing purity of the insulin.

Sometimes, however, patients continue to show these sensitization phenomena even when the purest insulin obtainable is used. In such cases insulin should be obtained from another source or desensitization methods, as applied to hay fever sufferers, should be tried. There may be, however, a residual group who will respond to any hypodermatic medication, protein or not, with phenomena simulating the allergic type. These cases, which Professor Macleod is inclined to regard as a sensitized reflex rather than true allergy, will probably be merely of a local nature. Thus far we have not had to deal with such a case. Williams (72) has reported a case of sensitization to insulin with very marked symptoms—swelling of the lips, urticaria, wheals and very marked weakness and prolonged prostration, and finally, exfoliation of the skin occurring on the substitution of one make of insulin for another. The lungs and heart were not

affected. Continuance of the insulin originally used has not induced sensitivity to beef insulin.

Geyelin (74) mentions mild serum sickness in 3 children, with successful desensitization to the insulin in 2 cases; the third apparently being very mild and clearing up spontaneously. Wilder (45) and his associates record severe protein reactions with 3 cases treated with the earlier preparations: urticarial rashes, nausea, vomiting, epigastric pain, diarrhea and profuse sweating are noted, together with reddening of the eyes, puffiness of the face and alterations in pulse volume and heart rate. As illustrating the idiosyncrasy of patients one of Wilder's cases received a small dose of a certain preparation which caused severe anaphylactic symptoms. We employed 120 cc. of the same solution, which was of low potency, intravenously in one dose in a coma case without deleterious effect. The patient recovered. Joslin (71) also mentions four protein reactions. None of our cases have had protein reactions so severe as those noted above. Their possible occurrence, however, should be remembered by anyone becoming responsible for insulin treatment in a diabetic.

Carbohydrate equivalent of insulin

The undoubted value of insulin as a therapeutic agent has led to attempts at assaying the unit in terms of carbohydrate metabolized. This problem was first attacked from the clinical side since its practical importance is so great. The most illuminating and valuable work, however, is that done in Professor Macleod's laboratory by Dr. Frank Allan (43), working with depancreatized animals. This has already been fully discussed by Professor Macleod in the preceding pages. Particularly suggestive is the formula developed since it corresponds to the law of enzyme action. Also the form of the curve helps to explain the clinical results obtained since all those presenting data on the subject are making use of a very similar type of diet, while those denying the possibility have employed most erratic dietetic procedures. That it makes a difference from whence the sugar is derived, all will agree. Wilder (45) has specifically stated that strict adherence to low protein diets increases the reliability of the results.

On *a priori* grounds the appraisal of the carbohydrate equivalent of insulin in clinical cases can never be wholly satisfactory. The patient, unlike a totally depancreatized animal, may possibly show fluctuations in the endogenous insulin production from time to time. Extraneous factors also play a part. Early or mild cases are not dependable. Desugarization may result in a rising insulin production. Woodyatt (46) selected cases with long metabolic records with relatively fixed tolerance and placed them on regimes which permitted the steady excretion of 20 to 30 grams of glucose daily. The insulin was then administered and the decrease in glucose excreted divided by the number of units gave the burning power of insulin per unit. A confirmatory test, discontinuing the insulin and determining the glucose excretion, was then performed to make sure there was no increased endogenous insulin production.

Wilder (45) also used selected patients, free from infections or other complications, placed on a basal maintenance food mixture. Average results of several days were used. The patients always showed appreciable glycosuria. The basal tolerance was determined by subtracting the amount of glucose excreted from the total glucose value of the diet and the diet then increased a definite amount for several days until sugar and nitrogen output became reasonably uniform. Insulin was then administered and the amount of additional glucose metabolized credited to the insulin administered. According to this method the unit assays between 1 and 1.5.

The precautions suggested by the above writers in determining carbohydrate equivalents are well taken. In Toronto the problem has been approached a little differently though the results are of the same general order. In fact when allowance is made for the fact that the insulin used by these workers (Iletin) was of lower unit value than that used in Toronto, Wilder's (45) results are the same as Campbell and Fletcher's (80). Woodyatt's (46) results are a little lower but this may be explained by the fact that he makes use of 1 gram of protein per kilogram of body weight, whereas both Wilder and ourselves have used two-thirds of a gram of protein per kilogram body weight. Dr. Frank Allan's results seem to indicate that a lower carbohydrate equivalent would result when protein constitutes a high proportion of the total calories.

The carbohydrate equivalent of insulin seemed to us meaningless except from the therapeutic standpoint in relation to a particular type of treatment. As, in Toronto, the abolition of glycosuria and the maintenance of normal blood sugar was regarded as an essential in adequate treatment of the disease it seemed of value to learn the power of insulin to induce glucose combustion under these conditions. It was early noted that marked arteriosclerotics gave discordant values. Infection in a diabetic, the presence of acidosis, of emaciation or dehydration, also high thresholds of excretion for glucose were found to disqualify patients for testing purposes. To avoid inclusion of the effect of a spontaneously rising tolerance for carbohydrate it is necessary to keep the patient a sufficient period of time on a definite diet to show that this factor is absent. The diet was increased and the amount of insulin required to balance the extra carbohydrate and to bring the blood sugar to the same level then determined. The total extra glucose divided by units of insulin then corresponds to the carbohydrate equivalent of insulin. In applying this method, however, it was found that the result was practically identical with that obtained by dividing the difference between the tolerance for carbohydrate on a basal diet and the total glucose of the diet (the number of grams of glucose excreted) by the number of units of insulin used to maintain the patient sugar-free. The results, using batches of insulin with assay of which we were reasonably sure, gave an average value for the carbohydrate equivalent of 2.2 grams per unit. In a series of fifty-three mild diabetics Joslin (81) found the average carbohydrate equivalent to be 2 grams per unit (Joslin, ed. iii., p. 41). Major (82) also finds the same value.

The clinical significance of knowing the glucose equivalent lies in the fact that most patients treated on the usual types of diet can be given a dose of insulin calculated so as to make them aglycosuric and can then be maintained in this condition by a somewhat smaller dose. Thus, the average number of grams of glucose excreted on a diet supplying the basal requirement in calories divided by 1.5³

³ This factor is derived from experience. It may be suggested that the excess insulin (50 per cent) over and above the carbohydrate equivalent of the glucose excreted is used to metabolize the excess blood and tissue sugar. Suppose the blood sugar in a 60 kgm. diabetic is 0.3 per cent, i.e., 0.2 per cent above the normal. The tissue sugar

gives the dose of insulin required for rendering the patient aglycosuric.

$\frac{\text{Grams glucose excreted}}{2.2}$ gives the average dose of insulin required to

maintain the patient sugar-free in the absence of other complicating factors. By application of these principles much of the usual, prolonged and elaborate methods of treatment become unnecessary. Certain cases when treated according to these principles may become sugar-free but not attain suitable fasting blood sugar levels, and it becomes necessary to add two units of insulin to the daily dose for each 0.01 per cent if it is desired to lower the blood sugar. A variation in this rule is useful in judging the increase in diet allowable in a patient on dietetic treatment. If, for instance, the blood sugar level is 0.08 per cent it will not become excessively high (over 0.12 per cent) if 10 grams of carbs and 40 grams of fat are added. (The total glucose in such an increase is $10 + 10$ per cent of $40 = 14$ grams; $\frac{14}{2} =$ seven units required to metabolise the increase; $\frac{7}{2} = 0.035$ per cent = maximum increase in blood sugar produced by the addition to the diet.

Sherrill (84), in a long paper, assembles evidence to demonstrate that, under widely varying conditions, the carbohydrate equivalent of insulin shows very marked differences not only in different patients but also in the same patient under different conditions. He also states that glycosuria and insulin requirement are governed to a very important degree by the total caloric value of the diet. No especially powerful glycosuric influence of protein, either because of its specific dynamic action or supposed toxic action, was found.

Frederick M. Allen (85) also discusses the insulin requirements under various conditions, particularly the amount of fat used and the total calories in the diet. He believes that the insulin requirement of the organism is governed by carbohydrate, fat and all other elements entering into the diet or metabolism. The need for insulin is related quantitatively to the body mass as well as the amount of food to

is somewhat less than this level (according to the work of Palmer (83)), say 0.1 per cent above normal. In such an instance the body would contain 60 gms. of sugar, requiring insulin for its metabolism. There are, however, many other possibilities open; e.g., the endogenous production of insulin may decrease when the stimulation due to the overload is removed. In any case the dose of insulin must be promptly reduced to a lower value when the patient becomes sugar-free.

be metabolized. These observations support the belief that insulin is the true internal secretion of the pancreas. The under-nutrition treatment of diabetes as opposed to other dietary procedures, is held to be justified. Credit must be given Allen and Sherrill for assembling such a mass of data showing the variation in carbohydrate equivalent under different conditions. It is, in a way, confirmatory of the work of Frank Allan, referred to above. As the carbohydrate equivalent of insulin in man was determined for the purpose of a systematic treatment of diabetes and it was recognized that varying the conditions altered or abolished its value, the polemical character of these papers seems to be unjustified. That the insulin requirement is related to body mass as well as the food to be metabolized is a broad deduction from the evidence presented. The interrelationship between the numerous factors with which insulin is associated will, it is hoped, become much clearer as the result of investigations now proceeding. In the meantime Allen and Sherrill's results are quite consistent with chemical laws, such as the mass law and the law of enzyme action.

EFFECTS OF INSULIN ON METABOLISM

Many investigators have contributed information of value in regard to the alterations of the metabolism of diabetics under insulin. Interpretation of some of the results is by no means simple in the present state of our knowledge and it is not assisted by the fact that some investigators have employed their results mainly as arguments for some particular theory regarding the cause or treatment of diabetes.

Respiratory metabolism

Particularly interesting are the results on the respiratory metabolism, though these must be interpreted with the greatest care. So many fallacies arise in this work that, in all probability, we must await the results of a prolonged study of the complete metabolism of a severe diabetic in a chamber calorimeter before all details can be elucidated. Quite apart from technical errors of analysis, such details as the physical condition of the patient, over-breathing, the presence or absence of acidosis, dehydration, infections or other

complications, the duration and severity of the disease, the protein, fat and carbohydrate stores of the body, together with the influence of food, will have a marked influence on the result. Without doubt, some of the discrepancies noted are due to these factors. The psychic factor is so obviously important that all investigators have endeavoured to rule it out, but the fact remains that insulin causes such a marked change in the mental state and the physical vigor of the patient that it is difficult entirely to accomplish this.

Some preliminary observations made in Toronto (30) on severe diabetics, showed a rise in the respiratory quotient from 0.72 to 0.91 and 0.94 and similar values, after administration of insulin. This, alone with observations on diabetic dogs (35), was accepted as a part of the evidence that insulin actually causes combustion of carbohydrate in the diabetic organisms. Having established this fact, it was not considered advisable to continue further with the respiratory studies in Toronto since to do so on an adequate scale would have entailed a greater expenditure of time than seemed advisable in face of the many other problems that awaited investigation, especially since others investigating the action of insulin, such as Wilder, Boothby and Joslin, were in a more favorable position to prosecute work in this field.

Wilder, Boothby and their associates (45) investigated the behavior of the respiratory quotients of two diabetic patients when given glucose or levulose, with and without insulin. When glucose was given without insulin no rise in respiratory quotient occurred and, indeed, a slight fall is recorded in one of the cases. With levulose alone there was a distinct and immediate rise in the respiratory quotient though to a less extent than in normal persons.

With glucose and insulin a definite though delayed increase was observed in the respiratory quotient in one case; in another the increase was within the limits of experimental error. With levulose and insulin, on the other hand, the rise in the respiratory quotient is immediate and well-marked. This appears to indicate that levulose is more easily utilized by the diabetic organism than glucose and may be the explanation why levulose is so inefficient in controlling hypoglycemia (80). Wilder and Boothby suggest that the effect of insulin is to convert carbohydrate into an easily oxidizable and

polymerizable form and the ketose, levulose, is closer to this reactive form than is the aldose, glucose. The specific dynamic action of the two sugars is about the same with and without insulin; the levulose curve being the higher in both instances. No evidence of a permanent rise in the basal respiratory quotient was noted. The basal metabolism was also little changed.

Joslin, Gray and Root (71) report an average increase in basal metabolism of 9.4 per cent. Three out of eleven cases showed no increase—1 case showed 24 per cent increase—in basal metabolism. Using smaller doses of insulin than Wilder and his associates and administering a mixed meal of protein, fat and carbohydrate, Joslin found, in some instances, that the respiratory quotient rose to a very moderate extent and, in another instance, when successive doses of insulin were used, from 0.81 to 1.02. There was a marked increase in the basal metabolism accompanying this rise in respiratory quotient. In one instance the ingestion of levulose alone caused a greater rise in respiratory quotient than when insulin was administered at the same time. The high respiratory quotient, however, persisted longer in the experiment in which insulin was used. The basal respiratory quotients before and after a period of insulin treatment show increases so small as to bring them within the limits of experimental error.

Fitz, Murphy and Grant (73) report the effect of insulin on the respiratory quotient in 4 cases of diabetes. In all of them the respiratory quotient was definitely increased. In two cases the respiratory quotient was still comparatively high on the third day following injection of the insulin. This suggests the possible storage of carbohydrate in a form utilizable by the body or a prolonged action of the insulin. After fourteen days without insulin the respiratory quotient returned to 0.73—level characterizing severe diabetes. Basal metabolism was not markedly affected except in one case where the total calories per hour rose from 32 to 41.

McCann, Hannon and Dodd (86), in several cases show the initial decrease in the respiratory quotient when glucose is administered after insulin, as previously shown by Wilder and associates. As insulin treatment progressed it is interesting to see this peculiar fall give place to a distinct rise in the respiratory quotient.

Kellaway and Hughes (56) have contributed observations on the effect of insulin upon the metabolism in a normal person. A medical graduate volunteered as the subject. Two experiments were carried out with ten and eight rabbit doses of insulin respectively. Definite increase in the respiratory quotient occurred coincidently with the fall in blood sugar, as had previously been found in diabetes. The observed increase in oxygen consumption affords definite evidence of increased combustion of glucose. The authors contend that the rise in the respiratory quotient is due in part, to transformation of glucose into some complex that is poor in oxygen. Though it is not unlikely that this is sometimes the case, calculations based on their data fail to lend support to the hypothesis.

Lyman, Nicholls and McCann (57) determined the respiratory quotient and heat production in 5 normal men and 7 diabetic patients before and after insulin administration. In all cases the respiratory quotient rose and the heat production increased. Epinephrin caused a rise in the respiratory quotient and heat production possibly because of overventilation. Administration of the two drugs simultaneously, or following each other, on the whole produced a less variation from the basal values for respiratory quotient and heat production than when either was administered alone. The alterations in the metabolism of diabetic subjects were not as marked as in normal men.

Davies, Lambie, Lyon, Meakins and Robson (87) found a decrease in the respiratory quotient after insulin administration in a patient with acidosis and suggested that the destruction of blood ketones leaves alkali free to combine with some of the carbon dioxide produced. In a second case the respiratory quotient rose decidedly following the injection of insulin. The bicarbonate reserve was, in this case, nearer the normal than in the previous instance.

Rabinowitch (88) has published a table showing the effect of sixteen units of insulin on the respiratory quotient, the sugar, acetone and CO_2 combining power of the blood, together with the titrable acid *plus* ammonia and the glucose of the urine collected at short intervals, in a severe case of diabetes with acidosis. The respiratory quotient rose steadily from 0.724 to 0.804, coincidently with a fall in sugar and acetone bodies and an increase in the CO_2 combining power of the blood. The excretion of sugar and acid decreased in the urine.

Since the respiratory quotients for diacetic acid and β -oxybutyric acid are 1 and 0.89 respectively, Rabinowitch is careful to point out that the observed rise in respiratory quotient may have been partly due to the combustion of the acetone bodies. He also groups diabetic patients according to their tolerance for total glucose (G), and shows that the basal respiratory quotient rises with increasing tolerance. In a diabetic, a tolerance of 75 grams G was increased by the administration of insulin to 125 to 150 grams G. The basal respiratory quotients during this period rose from 0.75 to 0.79, the latter corresponding to a tolerance of about 175 grams G. This difference has been interpreted as indicating storage of carbohydrate. Coincident respiratory quotients and blood sugar curves after the administration of glucose with and without insulin have been interpreted in the same way. Even in normals we know so very little about the blood sugar curve after glucose ingestion, though much work has been done on it, that one would be very reluctant to subscribe to the interpretation unless supported by a very large number of observations.

Burgess, Campbell, Osman, Payne and Poulton (89) found that a severe diabetic under treatment for some months with insulin (75 units per day) showed a very high respiratory quotient—1.19, 1.08, 0.98 on three successive days. Twenty-three and a half hours after the last dose was given the respiratory quotient was 0.86 and 0.86, and the following day it was found to be 0.84. In 1 case 100 units of insulin were given in error, and it was necessary to give 110 gram of carbohydrate to counteract its effect. It was found that 50 grams more sugar should have been excreted than actually was the case. A storage of $50 + 110 = 160$ grams of carbohydrate occurred under the influence of the insulin. On the third day the blood sugar rose, and 85 grams extra sugar were excreted. From these premises, they conclude that these 85 grams of carbohydrate have been stored in the tissues in some complex form and were then liberated as glucose. The remaining 75 grams of sugar they assume were burned.

On the whole, then, the evidence goes to show, in spite of the very evident difficulties of interpretation of the results and the comparative paucity of experimental data, that there is a rise in the respiratory quotient after the administration of insulin. There, further, seems to be a tendency for the high respiratory quotient to persist over

several days, indicating either that the storage complex produced from carbohydrate by insulin requires no further insulin in its catabolism or that insulin has a more prolonged action than has previously been believed. That the result is not due to actual increase in the endogenous insulin production has not been satisfactorily shown, though this is most improbable. In several of the cases the insulin treatment was not sufficiently long to permit of such an improvement in the condition of the pancreas. Particularly in the severe cases with decidedly low basal metabolism, an increase in the latter occurs after insulin administration. Additional evidence is provided by Wilder and Joslin and their associates that levulose is somewhat easier to metabolize than glucose. Even though these results lack the advantage of prolonged observation there is considerable evidence from the clinical course of diabetics under insulin that they are in the main correct. The explanation of the respiratory quotients exceeding one is still somewhat obscure. The simplest explanation is that carbohydrate is reduced to fat. There is no good reason to suppose that diabetics under insulin cannot convert carbohydrate to fat or, at least, into some other complex that is poor in oxygen. Observations on the metabolic rate and respiratory quotient occasionally bring to light anomalous results which cannot be satisfactorily explained, and it is better to reserve judgment until a larger amount of data has been accumulated.

Glycosuria

Loss of glucose to the body is not the most serious defect in the metabolism of the diabetic patient. It is, however, a warning signal of the failure to metabolize sugar efficiently, and on this account is of importance. The original observations (30, 68) that sufficient insulin would control glycosuria in all cases of true diabetes mellitus has been substantiated by every writer who has used it intelligently—so many now as to preclude the possibility of mentioning them. In approximately 500 cases of diabetes under treatment in Toronto insulin has not failed. Further, as Woodyatt (46) pointed out, a test with insulin is of value in the differential diagnosis of the glycosurias—diabetic, true renal and otherwise.

Insulin will increase carbohydrate utilization whether glycosuria is present or not and this has led some to ignore the well known fact that the diabetic showing glycosuria steadily and surely decreases in tolerance for carbohydrate. The other beneficial effects of the use of insulin, such as increased strength and vigor, brighter mentality, increase in weight, etc., may all be present but it surely cannot be satisfactory treatment not to give every chance to permit the patient's remaining tolerance to increase. Nor is it true that it can be *satisfactorily* replaced with insulin. Insulin is a crutch which slips at times. The larger the dose the greater the danger of hypoglycemia and the greater the difficulty of adjusting suitable dosage.

Admittedly, when insulin was first employed it was somewhat difficult to keep the patient aglycosuric. There was, therefore, some justification for those who, rather than risk hypoglycemia with preparations of insulin which varied exceedingly in potency, were content to obtain the other benefits of the drug for their patient. We hold the position, however, that glycosuria in diabetes is a failure in treatment and so have consistently endeavoured to avoid it with rather remarkable success from the outset. Frequent repeated blood sugar examinations have enabled us to test the comparative efficiency of a particular batch of insulin on a well stabilized patient and to judge of its relative efficiency for other patients. For smaller clinics or for individual patients this would have been impossible but today the preparations are so well standardized that hypoglycemia from this cause does not occur.

Wilder (45) and his associates designedly permitted glycosuria for the reason given above. Woodyatt has found it possible to keep his patients sugar-free in most cases. "Old cases with low fixed tolerance limits have been difficult to keep sugar free . . . and it would not seem necessary that they do so, since they have no natural tolerance to lose." Up to the dashes the writer would agree and, if it be true that they have no natural tolerance, would agree also to the latter part. Cases without tolerance are comparatively few in number and very difficult to treat.

Geyelin (74), working with children and encountering the special difficulties of feeding and caring for these little patients regarded it as

safer to permit glycosuria than to risk hypoglycemia. With the present more accurate assay of insulin he has recently adopted a policy of keeping the patient aglycosuric (personal communication).

Fitz, Murphy and Grant (73) report patients made sugar-free with insulin but give no indication of their general policy.

Allen (75) consistently endeavours to prevent glycosuria in his diabetic patients, whether under insulin treatment or not, and Joslin (81) specifically states, "A sugar free urine is just as necessary, in my opinion, with diabetics treated with insulin as diabetic cases treated without insulin. I believe that, unless the urine is kept sugar free during the additional length of life these patients will enjoy complications due to hyperglycemia, and glycosuria will ensue."

Hyperglycemia

The maintenance of normal blood sugar levels is not so easy in severe diabetics under insulin treatment as it is in the under-nutrition types of treatment, since the immediate, or blood sugar lowering, effect of insulin is of comparatively short duration and repeated injections must be used to accomplish this object. Fortunately it seems to become easier as the state of nutrition of the patient is improved and the tendency to hypoglycemia becomes less, possibly because of the increased glycogen content of the liver and muscle tissues. Campbell and Fletcher (80) observed in such patients, as well as in normals and non-diabetics, that a suitable dose of insulin would lower the blood sugar to the normal level for many hours without the occurrence of hypoglycemia. This is more difficult to accomplish in the emaciated subject and also in the patient who has an extremely low tolerance for total glucose. In certain of these cases hypoglycemia is very readily produced by small doses of insulin. Caution must be observed in commencing treatment in such cases.

Fortunately, even these cases are likely to have a high blood sugar and consequently a large amount of carbohydrate stored in the tissues as glucose, unless partial starvation has been invoked as a preliminary to insulin treatment. One can usually depend upon the tissue sugar to guard the patient against hypoglycemia. The dose of insulin used for freeing the urine of sugar must, however, be promptly reduced if it

rapidly becomes sugar free. In order to determine this, as well as the proper spacing of the dose, patients should have the urine collected at short intervals and tested for sugar, at least during the first forty-eight hours after starting insulin treatment.

These difficulties plus the additional expense and inconvenience of having blood sugar tests made have induced many to question the necessity of maintaining normal blood sugars in treating these severe cases. Those whose convictions permit glycosuria in the diabetic naturally have no trouble over this refinement of diabetic treatment. To those who feel that it has been satisfactorily demonstrated that the aglycosuric diabetic lives longer and more comfortably than if glycosuria is permitted, the question of permitting hyperglycemia is a very important one. The Rev. Mr. X acquired diabetes at twenty-eight, with typical acute onset. He lived thirty-two years without treatment and was not known to be sugar free for any time. A good many patients who knew him have not lived nearly so long. When I was called to see him he had gangrene of the anterior half of the right foot with a spreading infection in the leg. No pulsation could be felt in the sclerosed vessels below the femoral artery on the left side. He was drowsy; breathing was a labor; and he was very weary. His temperature was 102°F. He died more of infection perhaps than of the diabetic coma which supervened. But, in the previous years, was he comfortable? For twelve years he had suffered from cramps in the legs on moving about. He was ataxic; he was uncertain of the position of his feet; later he could not write because of cramps in the hands. His eyesight partially failed him. He lost flesh. Intermittent polyuria became continuous and coupled with enlarged prostate, incontinence developed. He himself noticed his growing irritability and the development of an aphasia and realized its import.

Many times a similar picture develops more rapidly. We know that in the normal person glycosuria does not occur and, further, that the blood sugar is never raised to any great degree even after the ingestion of large amounts of glucose. If insulin is used only to keep patients alive shall we in the future see more of these cases with marked degenerative phenomena? Not from the glycosuria surely, but hyperglycemia is the only other related symptom. If it be true, and it must be remembered that it is not yet proved, that hyperglycemia

predisposes to sclerosis of vessels and other degenerative phenomena, then sufficient insulin, endogenously produced or artificially administered, to keep the blood sugar normal is essential in treatment. But, further, we do know that glycosuria in the average case of diabetes is inimical to his best interests and that his tolerance for carbohydrate is more or less rapidly destroyed thereby. Consideration makes it apparent that it is not glycosuria, *per se* which produces this effect but excessive stimulation of the pancreas by the accompanying hyperglycemia, and it has been considered the best practice to remove this by diets below the tolerance of the patients. Those whose blood sugar could not be lowered by diet often broke to a lower level of tolerance. The pancreas of the aglycosuric diabetic who is allowed to have hyperglycemia is subject to the same overwork and resultant damage as the pancreas of the untreated patient. The results are possibly more gradual but there is no doubt as to the progressiveness of the condition.

Protein sparing action of carbohydrate

In a number of the cases reported by Banting, Campbell and Fletcher (69) the protein sparing action of glucose is demonstrated. Similar results have also been obtained by Wilder (45) and by Fitz, Murphy and Grant (73). Some of Allen's protocols (75) also illustrate this action. To show this action, all of these observers have depended upon the variations in output of urinary nitrogen in severe diabetics, with and without insulin. More complete metabolism experiments with analysed diets are highly desirable since there are several objections to this method with the type of diet commonly used by the diabetic patient. It is probable, however, from some preliminary observations that such a study will only increase the already striking evidence of the preferential utilization of carbohydrate over protein when sufficient insulin is available to burn the former. On the reduced diets commonly used in diabetic treatment this protein sparing action is of some considerable importance, since upon it depend the possibilities of the emaciated patient regaining necessary weight and strength, as well as restoring the functional efficiency of organs severely under-nourished. The mere increase in

weight of a patient, which may be brought about by retention of fat, salt and water, can have but a limited beneficial effect, and that mostly psychic in nature. The restoration of muscle and organ mass by actual increase in the total body protein is much more important for the continued well-being of the patient. The fact that such protein sparing action can be demonstrated on dietary protein allowances of $\frac{2}{3}$ gram of protein per kilogram body weight, as shown by Wilder (45) and ourselves (69), is an indication of the adequacy of such amounts of protein for the needs of the body.

Lipemia

Considerable theoretical interest is attached to the behavior of the blood fats under insulin treatment, but as yet they have been rather imperfectly studied in man. Quite early it was noted (30) that visible lipemia disappeared under treatment. Actual figures were not published but it did not appear more striking than one often observes on dietetic treatment alone. Davies, Lambie, Lyon, Meakins and Robson (87), in a preliminary communication, note qualitatively the disappearance of visible lipemia in 2 cases. Wilder and his associates (45) report analyses of blood fat in 3 cases without significant alterations. Joslin, Gray and Root (71) also report analyses showing a slow decrease in the blood fats. Considerable quantities of excess fat may be present in the serum without producing a visible lipemia. In our experience the total blood fats under insulin treatment tend to fall rather slowly. No definite relationship to insulin administered or increased carbohydrate metabolism has been made out. The disappearance of lipemia is certainly more rapid in those cases where no fat is being fed to the patient and acidosis is rapidly abolished. Hardy (103) has reported disappearance of lipemia retinalis under insulin treatment. Major (104) has shown that in two cases the eruption of diabetics due to deposition of cholesterol crystals and known as Xanthoma diabetorum disappears after treatment of the patient with insulin. He believes that the clearing up of the lesions is due to good blood supply and to disappearance of lipemia and hyper-cholesterinemia.

Body weight

Increase in the body weight of diabetics to any desired level can now be induced under insulin treatment. It should be pointed out, however, that in this case as in so many others, insulin merely makes possible for the severe case of diabetes a condition which is possible under suitable dietetic regimens in the patient afflicted with the milder forms of the disease. To many the evidence of the scales may seem a guarantee of progress but this is only true if one realizes the source of the weight change in the particular patient. We may dismiss at once the question of restoring to the obese diabetic the few pounds he has lost since there can be no question that overweight is not a favorable condition. The obese diabetic needs a fat reduction cure even more than the corpulent, middle-aged candidate for diabetes. In the former case the damage has been done but this is usually moderate in degree as far as tolerance for carbohydrate is concerned. Reduction in weight increases the effective mass of active pancreatic islets relative to the size of the individual and thereby relatively increases the tolerance of the patient. The man of forty-five, 5 feet 10 inches tall, who is 40 kgm. over-weight requires, merely to live, 350 calories more than if he were of normal weight. In addition, of course, he must expend energy to carry the superfluous fat. Reduction in weight in such cases should not, of course, involve loss in muscle mass or muscular efficiency.

Probably the most spectacular increases in weight, following insulin, occur in patients suffering from marked emaciation, acidosis and dehydration. In these cases it is easy to see that the increase in weight cannot be due to deposition of protein since insufficient of this food principle is ingested. Since the deposition of fat in the body is accomplished in almost an anhydrous manner, only 10 per cent of its weight of water being required in the process, the weight increase cannot be due to fat which is also insufficient in amount. The deposition of carbohydrate in the body is accompanied by three times as much water. The limited possibilities of glycogen storage, however, make it unlikely that much more than a kilogram increase in weight can be expected from this source. An approximate balancing of the total fluid intake and output gives the clue, namely: that the weight

increase is largely due to water retention. The type of diet served the diabetic is rich in mineral salts and these assist in maintaining a normal osmotic pressure in the tissues and the diabetic quickly attains a plump appearance, while the tissue turgor is distinctly increased. As Allen and Joslin have remarked, this affects particularly the face of the patient, but the whole body is involved to a greater or less extent. If sodium bicarbonate is also used this tendency may become very pronounced. The amount of weight increase in emaciated patients due to stored fluid without edema may be quite astonishing. Twelve pounds is not an uncommon amount for such a diabetic to store, but the adult in good nutrition will probably store less fluid. Even here the weight increase attainable without edema may be considerable. In experiments on water drinking the writer found six pounds increase in weight after the diuresis, due to drinking ten litres of water, had ceased. This increase in weight persisted for several weeks without evidence of edema or change from the original values in either hemoglobin, red-cell count or blood pressure.

In some cases where the fluid intake of emaciated patients was not sufficiently controlled edema developed. In ambulant patients this has been most striking and is due to change in position of the excess body fluid rather than in excess intake of fluid immediately before. In twelve hours a patient may lose the plump facies and become pinched, drawn and emaciated while examination reveals marked edema of the ankles and legs. The reverse change occurs in dehydration. This change in the condition of the tissues is not wholly due to a true loss of body fluid but may occur fairly rapidly when no urine is being secreted, and in spite of moderate fluid intake.

True increases in body tissue are acquired comparatively slowly. Emaciated patients fed with fat in excess of their needs may indeed store it but it is questionable whether increase in weight due to an inactive substance like fat is desirable beyond a certain point. An increase in total muscle mass or other tissue containing protein serves a useful purpose. The supply of essential body fluid to replace that lost is also desirable but it is important to realize that fat and excess fluids merely contribute a cosmetic effect and require energy to keep them warm as well as to move them about. The patient whose body

weight is between normal and 15 per cent below normal is the most satisfactory risk.

It may not be amiss to note that in the opinion of the writer, the phrase "insulin edema," recently encountered, is a misnomer, as it relates to a condition possible of attainment under certain conditions in patients on dietetic treatment alone. Further, we have yet to see the edema of a diabetic which would not disappear with salt and water restriction. If insulin is necessary for treatment, patients should not have it discontinued because of the development of edema.

Luxury use of insulin

The so-called "luxury use" of insulin was a matter of some concern during the earlier period of treatment with the drug, while supplies were still relatively scarce. At that time it was considered justifiable to place a limit upon the amount of insulin obtainable by any patient in order that others should not suffer. The large supply of insulin now available has brought about its use by many physicians who, being more or less unfamiliar with the clinical course of diabetes, are using it unnecessarily (luxury use). The use of insulin and precariously high diets to fatten a diabetic unduly, or to satisfy a gluttonous appetite, or to avoid the necessity of dieting reveals a lack of intelligent foresight on the part of the physician as well as a lack of resourcefulness in the treatment of his patients. For the patient himself there is increased danger of acidosis, with the constant risk of an incorrect balancing of the diet in relationship to insulin, so that either hypoglycemia or hyperglycemia and glycosuria are often present, resulting in decreased tolerance and degenerative phenomena associated with diabetes of long duration.

Several writers (81, 105, 106) have emphasized the danger of acidosis and coma resulting from the sudden discontinuance of insulin treatment. The result is not of course comparable with the prohibition of narcotics to the drug addict but is analogous to the results of sudden decrease of tolerance for carbohydrate in diabetic patients who acquire acute infections. Wilder mentions a patient who died consequent upon being without insulin for 36 hours. All patients

should be warned of this danger which is the greater the more liberal the diet and the higher the dosage of insulin ordered. The danger is probably increased by any exertion or by an artificially induced or artificially maintained state of hypernutrition of the individual. Were the so-called "cures" (temporary overfeeding of mild diabetics with accompanying insulin administration) now being attempted in Europe and elsewhere not unsound on other grounds this risk should act as a deterrent. Bed rest, increased fluid intake and immediate reduction in diet, particularly the fat, should be enjoined on a patient compelled to forgo insulin temporarily for any reason.

Polyuria

The increased amount of urine secreted by patients suffering from diabetes mellitus has usually been thought to be due to the diuretic effect of the sugar excreted along with it. Three factors—increased volume, sugar content, and high specific gravity—are commonly associated. Frequently a necessary uranalysis has been neglected because of a report of the excretion of a normal daily volume of urine. The fact is: the percentage of sugar in the urine may vary quite independently of the volume or the specific gravity and, further, on account of the variations in the amount of inorganic salts and other organic substances in the urine there is no way of even roughly approximating the urinary glucose output by the specific gravity or by any of the qualitative tests. In the majority of patients with diabetes amenable to dietetic measures there is marked reduction in the excessive thirst and polyuria when the patient becomes aglycosuric.

An unbroken night's rest and increased comfort during the day, when polyuria was reduced, have often induced a discouraged patient to persevere with dietetic treatment. All patients heretofore made sugar free by dietetic means did not receive an equal degree of relief from the polyuria and this lack of relief was ascribed to the diuretic action of the hyperglycemia, though other patients with fully as high blood sugar levels early became free from polyuria. The advent of insulin has not changed this situation. Some patients who could not be made aglycosuric without insulin show a decreased urinary output when the glycosuria is prevented by the use of insulin. Some eight

of our patients receiving insulin treatment, nearly all cases of the most severe type, continue to have polyuria in spite of becoming aglycosuric and having a normal blood sugar level. Polyuria in these cases appears to be an obligatory condition unrelated to diet or to renal disease. It is interesting to note that pituitary extract to some extent controls the polyuria, though it would be premature at present to postulate a combined pancreatic and pituitary lesion without further study of the condition. The power of the kidney to concentrate the urine is not lost in these cases since the specific gravity promptly increases under the influence of pituitary extract.

Duration of treatment

Evidence related to the time a diabetic is under treatment with insulin must be added to the above mentioned immediate effect. It is conceivable that a patient might show temporary improvement without permanent benefit from insulin or from any other type of treatment. Incidentally, it may be pointed out that the benefits derived from prolonged treatment with insulin furnish a further argument that it is the necessary hormone that is lacking in the diabetic organism.

Two years of treatment of severe diabetic patients with insulin, resulting in marked improvement in the physical condition of the patients and their ability to do work, together with a lack of any evidence of deleterious effects, is sufficient guarantee that insulin can be continuously used without its value becoming impaired by unforeseen occurrences. Under older methods of treatment these patients, or at least a large proportion of them, would now be dead. It is common experience that life expectancy in severe diabetes has been prolonged. The types of dietetic treatment employed by various clinicians have been so diverse that the use of insulin in treatment appears to be the only additional common factor to which the increased duration of life can be related.

The amount of food consumed by the diabetic patient under insulin treatment has been far in excess of his previous tolerance. The assimilation of these foods without glycosuria or acetonuria makes it apparent that they must have been stored or metabolized. The former possibility is ruled out by the fact that the body weight does not

become excessively raised, which would be the case if storage were solely concerned. Catabolism, of the foodstuffs, leading to greater energy output, accounts for the additional activities voluntarily undertaken by the patients. Since, previous to the use of insulin, all cases of severe diabetes were metabolising high proportions of fat relatively to carbohydrate and, in most instances, were showing acetonuria, it follows that all the increased food assimilated as a result of insulin is not converted into fat and burned as such. If this were not the case the already excessive proportion of fat to carbohydrate metabolized would be increased and pronounced acidosis would certainly have resulted. Absence of acetonuria removes this possibility and makes it certain that sufficient additional carbohydrate has been burned to establish a safe ketogenic antiketogenic ratio.

HYPOGLYCEMIA

Although insulin possesses so many beneficent properties for the sufferer from diabetes mellitus it is not without drawbacks and contraindications. Some of these have been touched upon in the preceding pages. Indeed, its specific properties of influencing carbohydrate combustion, if used to excess, cannot fail to affect the well-being of the patient. It is important to remember that with insulin we are dealing with a most powerful drug, many times as toxic as strychnine. Its toxic action arises from the fact that, when present in excess, it inhibits the utilization of, or fixes in an unavailable form, the soluble carbohydrate of the body. This suggested the possibility that the body tissues might not only be deprived of sufficient food, but also that an acidosis might be produced from the attempt to utilize fat in the absence of the carbohydrate. Certain preliminary observations by Collip, in which he stated that insulin causes ketonuria in normal rabbits, seemed to support this view, but the well-known susceptibility of rabbits to changes in food supply renders the result somewhat doubtful.

The writer has seen three deaths from hypoglycemia on under-nutrition diets, and Joslin also has reported four cases treated by dietetic methods. Woodyatt and Williams have also noted similar cases, and it is quite probable that there have been many other cases.

As noted above, we early became aware from the animal experiments that a condition of hypoglycemia would result from overdosage of insulin. The clinical state of these animals was carefully studied and reported as far as possible, and fortunately before any human cases were encountered the more marked stages of the condition were known and the treatment by carbohydrate and by adrenalin were tested out and found to be effective. A review of this work by Professor Macleod appears in *Physiological Reviews*.

Investigation on animals naturally fell short of giving a complete picture of the symptoms accompanying hypoglycemia, since subjective symptoms were not available for study. A report of the general features of this condition was made in the *British Medical Journal*, January 6, 1922, and the subject was more completely discussed by Fletcher and Campbell (70) in the *Journal of Metabolic Research*, November-December number, 1922, and additional studies reported at the meeting of the Society for Clinical Investigation, May, 1922. (Cf. *Proceedings Society for Clinical Investigation*, Abstract *Journal American Medical Association*, lxxx, 1641.)

The grave danger to patients from an unnoticed or untreated hypoglycemic reaction makes it important that every patient should be thoroughly familiar with it. With each bottle of insulin an instruction sheet is sent out giving this information as well as pointing out the cure. The warning cannot be too earnestly repeated; and in our clinic it has been the practice to induce an hypoglycemic reaction in patients about to be discharged and who may not previously have experienced one during treatment. In such cases, of course, there is no necessity for pushing the reaction very far. The patient should experience the early signs and symptoms up to the sweating stage, and also be instructed as to the cure of the condition. This precaution has been considered unnecessary in other clinics and, though we felt justified in maintaining it while the assay of insulin was uncertain, the number of reactions encountered decreased decidedly with the improvements in the assay.

Two deaths from insulin hypoglycemia have been seen. The first, an habitual drunkard, who had been refused insulin treatment at Toronto General Hospital because of this danger, received insulin from another clinic. Having been slightly inebriated, he arose in

the morning with a distaste for food and omitted breakfast but injected thirty units of insulin subcutaneously. He transacted considerable business, walked considerably, hurried all morning in order to catch a train at midday. About noon he was brought as an emergency case to the Toronto General Hospital, having collapsed on the street. He was cyanosed; his heart was dilated; the heart sounds were weak; his breathing was difficult; he had edema of the lungs. Stimulants were given but he died before a needle could be boiled to administer glucose intravenously. The other case, from our own clinic, was a severe diabetic using forty units of insulin per day, who suffered from a gastrointestinal upset with vomiting. Without notifying his physician he took ten units of insulin without any food. Noticing the onset of the hypoglycemic reaction he ate an orange and, later, telephoned to his physician. Feeling improved he neglected to carry out the instructions given him to rest and take more glucose. A sudden collapse occurred followed by death before assistance arrived.

The symptoms and signs of the hypoglycemic state are more or less related to the level of blood sugar attained and become progressively worse as the blood sugar falls. In nearly all cases the blood sugar is below the normal level before the patient notices the abnormal symptoms. Occasional instances have occurred when symptoms that stimulate those due to hypoglycemia have been described while the patient's blood sugar level was still above 0.1 per cent (0.110 and 0.130 per cent). These have also been noted by others who have used insulin in treating their cases. It is possible that they may be explained as protein sensitization reactions but this is not certain. They apparently occur in patients whose blood sugar level is permanently above normal and who have high thresholds of excretion for sugar. This type of symptom is deserving of further study since the speed of change of the blood sugar level may be in some way the responsible factor, or it may be that hyperglycemia, in cases showing the symptom, is a necessary condition for the welfare of the individual, just as many arteriosclerotic patients feel better when their blood pressure is not lowered by means of drugs below a certain level.

The blood sugar level at which symptoms usually appear is about 0.08 per cent. It is known, however, that blood sugar percentages of 0.08 and even 0.06 per cent are many times encountered in normal

persons who have not been treated with insulin without the characteristic symptoms of hypoglycemia. Also reactions which occur long after the insulin has been given are much less acute, other things being equal, than those which occur earlier. In such cases few symptoms may occur although the blood sugar is very low, and then suddenly disorientation, confusion, and even unconsciousness supervene. It may possibly be that it is the rate of lowering of the blood sugar—the dynamic factor—rather than the level attained that is responsible for the differences. Accidentally in one most remarkable case a blood sugar of 0.032 per cent was found. The patient presented no symptoms or signs of reaction and he had been given his evening meal before the result of the blood sugar test was known. Less marked degrees of hypoglycemia without symptoms have been observed on a number of occasions. The lowest blood sugar we have noted is 0.025 per cent. With the administration of glucose this patient recovered.

There is, then, no absolute blood sugar level at which certain signs or symptoms invariably occur. Occasionally a symptom from the severe class appears at a high blood sugar level. Nevertheless in most cases one can predict the blood sugar level from the severity of the associated symptoms.

The earliest symptom which most patients describe is a vague, indefinite feeling of uneasiness, an uncomfortable sense of impending danger. A peculiar mask-like immobility of the face with dilation of the pupils is often seen. Marked hunger may be complained of. Increase in pulse rate, with weakness, pallor, flushing, nervousness and tremulousness are early evidence of hypoglycemia. The blood sugar at this time is usually between 0.08 and 0.07 per cent. With lowering of the blood sugar to 0.07 to 0.055 per cent nervousness gives place to anxiety and faintness; excitement and emotional disturbances with occasionally vertigo may occur. The physical signs at this stage include sweating, often most profuse, diplopia and incoördination. Marked excitement, emotional instability, sensory and motor aphasia, delirium, disorientation, delusions, confusion and bradycardia occur with blood sugar levels of 0.055 to 0.040 per cent.

With lower blood sugar levels the blood pressure may fall. Usually, however, it is well sustained. Unconsciousness has not been observed to occur until the blood sugar has reached 0.035 per cent. It may be

accompanied by hypotonicity or hypertonicity of muscles and either lost or increased reflexes. Tremor, hypertonia, individual muscle twitchings and marked mental irritability mark the recovery from unconsciousness. Convulsions in children have been reported by Geyelin (74) and by others. Up until two months ago they had not occurred in any of our patients. In this case the child had a few clonic twitchings, which had ceased before my arrival, being succeeded by unconsciousness. Glucose solution injected intravenously promptly removed the symptoms.

The direct cause of hypoglycemia is the administration of an excess of insulin relative to the carbohydrate then available. It may result from excessive amounts, or unsuitable frequency of dosage or unsuitable time of administration. Failure to absorb the carbohydrate of the diet because of vomiting, diarrhea, duodenal ulcer, pylorospasm, etc., may render an otherwise suitable dose of insulin excessive in amount. Over-exercise may deplete the carbohydrate stores of the body or cause the endogenous production of insulin to become increased and so render the dose excessive.

Treatment of the condition consists in increasing the amount of circulating glucose in the body. Noble and Macleod (90) have shown that, injected subcutaneously, glucose is the only satisfactory antidote for the condition in rabbits. In man, when administered by mouth, a more extensive list of substances are suitable. With the exception of maltose, which is nearly as efficient, Campbell and Fletcher (80) have found the most satisfactory restorative is glucose. Substances other than glucose probably act in accordance with the ease with which glucose can be obtained from them. The order of efficiency is: glucose, maltose, galactose, lactose, sucrose, levulose and glycerine. Starches are more slowly effective. Wilder (45) and his associates have confirmed the observation (68) that epinephrin may be used to combat insulin hypoglycemia in man and also the relatively short duration of its effect, and emphasized the necessity of administering carbohydrate immediately the patient is able to swallow. Burn (48) states that pituitrin is effective in animals. For several months we have considered it inadvisable to recommend these drugs for clinical use in hypoglycemia. They are needed only during

the unconscious state. The objections are both theoretical and practical but, in our experience, are not based on any deleterious effects attributable to the epinephrin itself. Certain cases, however, fail to show improvement with epinephrin because this substance acts by liberating glucose from the glycogen stores. Since these stores are much depleted in emaciated patients and may be very low after exercise the amount of glucose set free by epinephrin may be too small to relieve the hypoglycemia. At the same time it must be remembered that the work of the heart is interfered with by lack of available glucose. Since epinephrin increases the heart work the result might be most serious if no glycogen stores are available. Lyman, Nicholls and McCann (57) report 2 cases in which epinephrin failed to increase the blood sugar during insulin hypoglycemia.

The quantity of glucose necessary to restore the patient varies with the severity of the condition, that is: it is related to the degree of hypoglycemia present. Hence, the importance of determining by the clinical evidence presented the approximate blood sugar level. Though it will probably do little real harm it is better practice to avoid excessive sugar intake and the resulting glycosuria. The presence of glycosuria may make it difficult to interpret the clinical chart for two or three days and this symptom should be avoided if speedy treatment is desirable. If the premonitory symptoms of hypoglycemia are noted half to one orange will usually be effective. If the sweating stage is reached (blood sugar 0.07 to 0.055 per cent) the carbohydrate in an orange is insufficient unless a meal is imminent. A level tablespoonful of corn syrup is usually effective but should be repeated if relief is not experienced within ten minutes or if the patient recovers only temporarily. More severe cases should have three or more table-spoonful of corn syrup, or its equivalent, administered by mouth at once. Patients who are unconscious should be given an intravenous injection of glucose solution. Fifty grams or more of pure glucose may be necessary in these cases. Every insulin patient should be given ample opportunity to realize his danger from the unintelligent use of this drug.

SIMPLE KETOSIS, ACIDOSIS AND COMA

For our present views on these subjects we are largely indebted to the papers of Woodyatt (91), of Shaffer (92) and of Wilder (93).

Limitation of space prevents discussion here of some of the most important phases of intermediary metabolism most clearly set forth by these writers. It may be taken, however, that a definite maximal amount of fatty acid may be completely oxidised without acetone bodies (at least of any moment) being produced when a certain amount of glucose is being burned. This glucose (G of Woodyatt) may be derived from the carbohydrate in the food, from 58 per cent of the body protein and from 10 per cent of fat, since it is known that glycerine (10 per cent by weight of the fat) can produce an equal weight of glucose. The writer does not intend here to go into the question as to the absolute accuracy of these figures. The significant point is that they form the basis of a most successful method of treatment for diabetes.

Marked increase in the ratio of fat to glucose undergoing metabolism will cause the production of the acetone bodies—acetone, acetoacetic acid and β oxybutyric acid. In small amounts these substances may appear in the blood and urine without causing abnormal physical signs or symptoms. This condition may be called simple ketosis. With increased over-production, or delayed elimination of the acetone bodies, however, physical signs appear, due partly to disturbances in the hydrogen ion concentration of the blood and partly to the toxic action of these substances. Milder degrees of this toxic action constitute a state of acidosis but severe degrees, due to an increased amount or a prolonged action of these substances in the body, result in diabetic coma. With these symptoms are associated alterations in the mineral and water balance of the blood and tissues, and in all probability changes in the physical and chemical state of the body proteins. In cases in which severe acidosis and coma occur there is evidence of dehydration. Many times acidosis is due to a temporary lowering of tolerance resultant on an acute infection. In the severe stages it seems probable that the pancreatic damage is so severe that the islet cells have ceased to function. In the first patients treated with the pancreatic extracts (30) an immediate reduction in the ketonuria occurred and encouraged the belief that acidosis and coma might be efficiently treated by their use. It later was found (68, 34) that not only ketonuria and ketonaemia could be made to

disappear but also that the CO_2 combining power and alveolar CO_2 tension of the blood return to normal levels when insulin is used. Coincidentally with these effects the patients were relieved of the symptoms of acidosis and coma from which they were suffering. Nearly all our collaborators have taken occasion to confirm these results and there are now on record numerous tables illustrating these effects of insulin. More recently Cullen and Jonas (94) have demonstrated exceedingly low hydrogen ion concentrations in the blood of diabetic coma cases with subsequent return to normal values under insulin treatment.

The amount of insulin necessary to control acidosis varies considerably. It is well known that dietetic treatment will control the condition in some cases, and patients showing simple ketosis without clinical symptoms may usually be relieved by this means. The more severe degrees of ketosis may require insulin. Many patients exhibiting the severer degrees of ketosis would undoubtedly recover on suitable dietetic treatment, rest, warmth, etc., but when definite clinical symptoms are present it is preferable to use insulin than to risk the occurrence of coma. With the exception of cases of acidosis occurring during infections an initial dose of forty units of insulin and twenty units every four hours for six doses will be ample to control the situation. Most cases will require much less. The required dose of insulin to control acidosis during infections is much more difficult to approximate. The tolerance of insulin in this condition is enormous but the patient must be closely watched. In all cases carbohydrate should be supplied in amounts sufficient to prevent hypoglycemia, and the urine repeatedly tested for sugar. Suitable diet should be given as soon as the patient will take it and the insulin reduced gradually. Thirty to forty units should be given daily for two or three days until the danger of relapse is over. It is desirable also to observe the alveolar CO_2 tension, CO_2 combining power of the plasma and the blood sugar, but most cases can be successfully treated with urinary examinations for glucose and diacetic acid alone. The prompt treatment of acidosis with insulin when first diagnosed should diminish the number of coma cases seen in the hospital. Cases of coma about to be transferred to the hospital should also receive, as soon as possible, fifty

to sixty units of insulin with carbohydrates (by stomach tube if necessary).

There are very real benefits from dietetic treatment in cases of acidosis. We have not, however, cured diabetic coma by these means nor have we cured coma by the use of alkalies. The under-nutrition school regards it as impossible for coma to develop in patients on their type of treatment. Unfortunately this has not been our experience: on the contrary, even on starvation in cases without sufficient tolerance, coma may supervene. In this connection the experience of Fitz, Murphy and Grant (73) may be noted. Table VIII, page 764, in the *Journal of Metabolic Research*, volume ii, records a case on treatment for five days by Joslin's method, followed by two days of fasting. The acidosis increased. Small doses of insulin plus a diet relatively high in fat and total calories improved the acidosis. Joslin's (95) case 2366 developed coma in the hospital and recovered under "starvation" treatment without insulin or alkalies. This fact, and the fact that 15 other patients with acidosis recovered, should not, however, be used to discredit either the insulin or alkali therapy of diabetic coma. In 1921, while visiting the Mayo Clinic the writer saw Wilder treat an advanced diabetic coma with alkali. Recovery ensued and the patient was living and doing well a few months ago. During a visit to Toronto in 1922 Wilder saw Fletcher treat a child of six years in very severe acidosis, though still able to be roused, with insulin. This patient also recovered. At the time the American manufacturers of insulin were encountering great difficulty in maintaining the potency of the drug and Wilder, along with many others, was somewhat skeptical of its value. He expressed himself as satisfied that insulin was the effective agent in this case. The patient is still progressing favorably on dietetic treatment with insulin. None of these physicians however, failed to use other means of treatment. The other measures—rest, warmth, fluids,⁴ enemata and supportive

⁴ The influence of the dehydration factor in many of these cases is but little less important than the acidosis itself and its treatment by liberal fluid administration must not be neglected. Cases of coma due to dehydration alone, though fortunately rather uncommon, do occur. Much more commonly dehydration is present, often unnoticed, in various febrile diseases, conditions associated with vomiting and intense diarrhoea, the thyrotoxic crises and particularly in prolonged diabetic acidosis when the blunting of the sensation of thirst together with polyuria may give rise to a remarkable loss of body fluid.

measure applied to the circulation—are not needless; they are valuable adjuncts in treatment. It is the verdict of many investigators that the statement “Insulin is a specific in the treatment of diabetic coma” still stands, but today one would not care to treat coma by means of insulin alone.

With regard to the dose of insulin in coma and its administration, there is still a difference of opinion. The majority of investigators, including Joslin and Wilder, are in favor of small dosage. With the wide circulation of Joslin's (96) paper advising ten units of insulin as an initial dose in coma cases and the emphasis that has been placed on the dangers which may attend the treatment, 2 cases in one town, treated by a local practitioner, received five units of insulin each. Needless to say they died. Apparently Joslin has more recently changed his policy (95) and in case 3382 he gave forty units immediately, that is, four times as much as he previously recommended as an initial dose. This was followed by further injections so that one hundred units are given in six hours. We often give one hundred units immediately. Allen (75) and Woodyatt (46) use smaller initial doses (sixty to eighty units). We have not observed any deleterious effects from such large doses provided measures are taken to prevent hypoglycemia. For this purpose it has been the custom in Toronto to use 1 gram of glucose or other carbohydrate for each unit of insulin. Joslin regards this as excessively high, pointing out that a blood sugar of 0.5 per cent means 15 to 25 grams of sugar in the blood and that there may be other carbohydrate stores such as glycogen and protein. It is, of course, unnecessary to state that blood sugar is not always 0.5 per cent in coma and, indeed, may be normal. Further, it is not known that the glycogen stores use further insulin for their metabolism. The depancreatized dog may not develop a high DN ratio and low respiratory quotient and glycogen may be present in the liver for three days. The glycogen stores in the human diabetic in coma, in any event, must be rather small. The protein breakdown is small in amount and but little insulin will be used for metabolizing carbohydrate from this source. Moreover, glucose derived from this source is not so efficient an antiketogenic agent as that derived from carbohydrate itself. These imaginary safeguards should not, therefore, be relied upon to save a patient from an hypoglycemic death. The safest

practice is that a retention catheter should be placed in the bladder and the urine tested hourly for glucose and if this becomes low in amount more glucose should be administered immediately. But, further, it is well known that carbohydrate is burned in preference to fat and protein. By supplying this carbohydrate along with sufficient insulin to metabolize it no further breakdown of body fat or body protein is necessary to provide energy. Thus no further acetone bodies can be produced. The patient must then burn or excrete the stored ketones only in order to be relieved of his ketonic acidosis. Glucose is needed both to prevent hypoglycemia and to stop ketone production. The theoretical danger of damaging the pancreas through the production of hyperglycemia by administration of carbohydrate should not influence one for two reasons: the first being that the pancreas of a patient in coma is apparently inactive and the second that hyperglycemia is usually present in any event. It is interesting to note that Joslin (95) permits the latter as necessary to guard against death from hypoglycemia. In these views we have the support of Allen, who writes "This advice is practically identical with that already given by the Toronto investigators who are fully justified in their view that the harmfulness of carbohydrate excess for a day or two is negligible in an emergency of this kind" (75).

The first 15 coma patients treated in Toronto (30, 34) with insulin received no alkali. That the larger half of these cases recovered, sufficiently demonstrates the fact that insulin with other measures, as noted above, but without alkali, is effective in the treatment of coma. It is felt, moreover, that the results would have been much better had severe infections, such as pyemia and pneumonia, been absent in several of the cases. During the investigation of these and other cases of acidosis (34), it was found that in some instances the ketones could be made to disappear from the blood and urine without a corresponding increase in the CO_2 combining power of the plasma. This was attributed to a deficiency of base or unusual combinations of the remaining bases of the blood and tissues. In a paper read in April, 1923, Fitz, Bock, Starr and Field (97) noted the same condition but attributed it to the effect of an hitherto unidentified organic acid. More detailed evidence in favor of this view has recently been furnished by Starr and Fitz (107).

There is no doubt that a majority of coma patients have their CO_2 combining power raised to the lower limits of normal (50 volumes per cent) by the combustion of the ketonic acids under the influence of carbohydrate and insulin. In such cases (which, by the way, cannot be distinguished from the others except by prolonged chemical investigation) the use of alkali will not be necessary except in so far as it may be of value in shortening the period of coma and the abnormal chemical states associated with it. That this is worth while the majority of investigators are prepared to accept. In the other cases who present a low CO_2 combining power after the ketonic acids are burned, the use of alkali is imperative.

Palmer and Van Slyke (98) have shown that the administration of 1 gram of soda bicarbonate per 84 pounds body weight raises the CO_2 combining power of the individual's blood one volume per cent. Since, in most cases, we will be dealing with a patient whose CO_2 combining power will rise to fifty volumes per cent with the katabolism of the ketone bodies, it seems safe to use 20 grams of soda bicarbonate per 84 pounds of body weight, thus raising his CO_2 combining power to seventy volumes per cent—a value still below the upper normal limit—and alkalosis will be avoided. In these cases some acceleration in the improvement of the patient may be expected. But in the occasional case the ketones are destroyed, and insufficient alkali is thereby released to raise the CO_2 combining power. If the CO_2 combining power rises but to twenty-five volumes per cent that patient will probably still die of acidosis. In this instance the addition of twenty volumes per cent to the CO_2 combining power is a very decided factor in the relief of the patient. The amount of alkali necessary in the average case will be about 30 grams of sodium bicarbonate or its equivalent. Many other investigators have adopted a similar dosage of alkali. Should later determinations of the CO_2 combining power show insufficient alkali reserve more soda may be administered. The reaction of the urine should not be relied upon as a guide to alkali therapy in these cases. If the urine becomes alkaline the therapeutic dose may have been very markedly exceeded and the patient brought into danger by alkalosis.

With the use of alkali in any case, Joslin entirely disagrees, pointing out that 4 out of 5 cases do not need alkalies. Joslin's (95) question

is "Do you think there is any case of diabetes whose tissues are so robbed of sodium, potassium, calcium and magnesium, and whose kidneys are so devoid of the power to form ammonia that one ounce of soda will save him?" But, why did the coma develop if these substances were present in the body and available to save him? The quantity of soda used is not material to the argument so long as it is the therapeutic dose. If, indeed, alkali will only help to save one out of five comas, surely a 20 per cent less mortality in diabetic coma is acceptable, and it is worth giving it to all 5 cases in order to save the one.

THE GENERAL EFFECTS OF INSULIN

The physician has numerous criteria by which to judge the progress made in the treatment of the diabetic. Though it is desirable, in most instances, to teach patients as much as possible about their disease the patient himself is prone to judge his progress by his subjective sensations and the general condition of his body. With dietetic treatment in the less severe cases marked improvement was possible and some improvement nearly always occurred. With insulin the more severe diabetic whom previously we considered ourselves fortunate to be able to keep alive, has attained to a state of general health approaching the normal (68, 69). With increasing strength the patient not only loses the sense of weakness and continuous state of fatigue but muscle activity becomes a joy to him. Dryness and harshness of the skin and hair disappear. Skin infections clear. Allen (75) seems of the opinion that infections are more frequent in patients under insulin treatment than on under-nutrition diets. This may have been true in hospital practice if care was not taken to isolate diabetics from other persons having infections. It is most certainly not so in our experience in the home, nor in hospital practice provided the same care is exercised to prevent hyperglycemia as was the case when dietetic treatment alone was available.

The insatiable hunger of the emaciated, under-nourished, diabetic disappears with the improved state of nutrition, and is replaced by a healthy appetite for regular meals. We have found that the menstrual function has returned in several instances; so also, occasionally

has sexual desire. Eyesight usually improves in those cases with lesser degrees of local damage. The restoration of physical vigor is such that most patients are able to return to work. Most interesting is the change in the mental state of the patient. From depression, melancholia, inability to think, and often black despair, patients recover to an astounding degree their mental alertness, cheerfulness and interest in their surroundings. Irritability and insomnia disappear.

Previous to the use of insulin the carbohydrate tolerance of a patient was known to improve under suitable treatment. This was not at all dependent upon a mysterious beneficent influence of under-nutrition but upon adequate attention to the principle of rest to the damaged and overworked pancreas. It probably was true that in some patients with a low tolerance the pancreatic function was rested by under-nutrition and that their tolerance improved thereby. It cannot be too emphatically stated, however, that all grossly under-nourished patients did not derive this benefit and, consequently, were worse off than before the treatment was undertaken. On the other hand many patients, adequately nourished and in whom the blood sugar was carefully maintained around normal values, exhibited as marked increases in tolerance or endogenous insulin production as starved patients. Other cases, whether under-nourished or not, have developed a fixity of tolerance which is unaffected by treatment. The difference probably lies in the degrees of damage existing in the pancreas. In some cases the degenerative process may have reached a stage where healing takes place only by loss of functional tissue and in others partial restoration of damaged cells may have occurred.

Insulin provides for some patients an opportunity for resting the pancreatic function, which was impossible by other methods of treatment, and as a result increases in tolerance are not uncommonly observed. As the majority of the Toronto cases have been selected from the class of patients with long continued, severe diabetes it is natural that we have not seen so great a return of tolerance as other investigators. The proportion of patients with uncomplicated diabetes who find it possible to discontinue insulin after a time is comparatively small, but many show an increase in tolerance which though small is yet sufficient to render it possible to supply a working diet

with the same dose of insulin as was previously necessary when only a basal maintenance diet could be given. In some cases the insulin dosage may even be reduced. It seems possible that this so-called recovery of tolerance which usually occurs in the first months after treatment with insulin should be regarded rather as release of a latent tolerance submerged by previous overwork of the pancreas.

The question of an actual regeneration of islet tissue must be very cautiously approached. It can be settled only by time and it seems unfair to arouse hopes that are not justified by experience. In some of the mild cases and particularly in the young this is a possibility. It may be pointed out that children, who have not attained adult body weight, present the additional possibility that, under careful treatment and rest of the pancreas with insulin, a normal growth of the pancreas with increase in age may occur and thereby a greater tolerance for carbohydrate may be acquired. Close observation of some cases is being made to determine this point. Whether we are preventing diabetes in cases of acute pancreatitis with glycosuria and acidosis by administering insulin and reducing the diet it is impossible to state with any certainty. Some of the cases, before the days of insulin, recovered on a reduced diet, while others became true diabetics. An interesting and unexpected development in four cases of pancreatic with glycosuria was the marked reduction of pain after the administration of insulin. Further observations are necessary.

Influence of age

All patients using insulin do not receive equal benefit from its use. In part this is dependent on the severity of the disease and on various complicating factors, but there is also an influence which seems most directly related to the age of the patient. The most satisfactory results are obtained in young adults, the middle aged severe diabetics, and children over ten years. The aged patient generally shows a much less rapid recovery of strength, gains weight more slowly, and he requires a proportionately larger dose of insulin to produce a given amount of improvement. Dietetic treatment is more difficult in the young child because of greater liability to gastrointestinal upsets, and a tendency to refuse food. The collection of urine is more

uncertain and blood sugar determinations are more difficult to obtain. Some of the above difficulties are likewise encountered when the patient is treated by dietetic methods alone. When in addition the periodic administration of insulin is required by an aged individual or a very young patient the maintenance of a normal blood sugar level becomes decidedly more difficult.

Aged diabetics also suffer from many complications in part related to their age and also contributed to by the diabetic state. Coldness, numb extremities, muscle cramps, parasthesia, neuritis, ataxia, incoördination, etc., are dependent usually upon permanent organic changes in the circulatory and nervous systems, and the possibilities of improvement under insulin are rather remote.

Insulin in conditions other than Diabetes

Woodyatt (46) suggests the use of insulin as an aid for differentiating glycosurias of diabetic origin from that of renal origin or arising from other causes. In the renal cases where the glycosuria is small but continuous irrespective of diet, we have found, in agreement with Woodyatt, that if the blood sugar level is low insulin in doses several times as great as should stop the glycosuria has no appreciable effect. Fletcher has found that the hyperglycemia of arthritis responds to insulin. Of more purely physiological interest is the observation of Hepburn and Campbell that the hyperglycemia and slight glycosuria of carbon monoxide poisoning in man is promptly controlled by insulin. This effect of insulin was previously observed by Banting, Best, Collip, Macleod and Noble (58) working on rabbits.

The use of insulin in the pernicious vomiting of pregnancy is, in our experience, unnecessary. No case of post-operative acidosis has required it and it is difficult to see what purpose it could serve. Its use in the non-ketonic acidoses is even more questionable, and indeed one may be permitted to doubt its value in conditions other than those in which there is a distinct deficiency of endogenous insulin production. Those who insist upon regarding all the ills that flesh is heir to as due to an indefinite endocrine unbalance will do well to pause before applying insulin recklessly. Insulin is a powerful drug.

Insulin in Diabetes in Pregnancy

Diabetes in pregnancy presents a peculiar problem. In many cases not one but two metabolic defects exist: one, the diabetic condition, and the other a very marked disturbance of the ketogenic-antiketogenic ratio consequent upon the damage produced in the liver by pregnancy. In all normal pregnancies there is a deficiency of available base and a reduced alveolar CO_2 tension, indicating a liability to acidosis from non-ketogenic substances and, in addition, there is inability to use as much fat, relative to total glucose, as in the non-pregnant state. As a rule, when these patients are on an ordinary mixed diet or one that is relatively high in carbohydrate the ketogenic substances, since they are formed in but small amounts, do not contribute much to the acidosis. In a proportion of cases, however, more severe liver damage exists and considerable amounts of acetone bodies appear in the blood and the urine. Harding, in Toronto, has shown that most of these cases can be controlled satisfactorily by administering glucose and an adequate amount of fluids. Thalheimer (99) suggested the use of insulin in addition to glucose to control the condition in severe cases. It is difficult to appreciate the use of the drug in this type of case. In Toronto no case of uncomplicated pernicious vomiting sufficiently severe to warrant the trial of insulin in addition to fluids and glucose has been encountered in the last two years.

Pregnancy in diabetics, or the development of true diabetes mellitus during pregnancy is especially prone to be accompanied by liver damage with resultant failure to burn completely the acetone bodies and, probably, also a relative inability to produce glycogen when the carbohydrate is imperfectly metabolized. Particular care is necessary to avoid acidosis in these cases and when it becomes established the acidosis is decidedly more difficult to control—marked nausea and vomiting being prominent symptoms. Since pregnancy during diabetes tends, as a rule, to increase the severity of the patient's diabetic condition both during and, usually, after the pregnancy it is inadvisable for the diabetic to become pregnant. The high mortality of the fetus and young infant also renders childbearing in diabetes hardly worth the risk to the patient. Once established, however,

the question often arises as to whether the pregnancy should be terminated or not. Joslin (81) seems to feel that it should not, and I would agree with him so long as it does not involve undue risk to the mother. In some cases induced labor a short time before term, particularly in primiparae, seems to me justifiable, as in well conducted institutions it has little influence on the fetal mortality and saves the mother a prolonged parturition during which she is particularly liable to develop acidosis. Lactation should be avoided since it is a severe drain on the maternal metabolism.

The dietetic treatment in the pregnant diabetic is complicated by the necessity of using a larger relative proportion of glucose to fatty acid so as to avoid acidosis. This should be instituted at the outset of treatment and if the insulin production of the patient is insufficient to maintain an aglycosuric state, with a normal fasting blood sugar, more insulin should be supplied artificially. The total glucose of the diet in grams should at least equal the fat. The protein allowance should not be excessive—1 to 1.25 grams per kilogram of body weight is sufficient. The total energy of the diet should be maintained comparatively lower than in the nonpregnant woman and the patient, as is usual during pregnancy, should be taught to rest as much as possible. In many pregnant diabetics such a plan of treatment is necessary to avoid acidosis, the symptoms of which very much resemble and are, at the same time, more serious than the pernicious vomiting of pregnancy. Insulin will be found necessary to carry out this program, and it should be noted that following parturition some patients may with advantage revert to a diet somewhat higher in fat and lower in total glucose with omission or reduction of the dose of insulin used.

Notwithstanding the use of treatment ordinarily effective, or perhaps because severe acidosis is already present when the patient is first seen, the condition of the patient may become progressively worse. With continued vomiting, loss of food and fluid, the patient grows steadily weaker and more dehydrated. Such patients exhibit a most distressing and dangerous degree of acidosis and may readily lapse into coma. Insulin should be pushed, remembering that time lost in the treatment of such cases cannot be made up later by larger dosage of the drug. Insulin used immediately will control the acidosis but if this is very severe and vomiting cannot be checked it may

be inferred that severe liver damage has been done and that the condition will readily recur. In the more severe cases it becomes obviously impossible to carry the patient to term on an emergency basis. Sometimes such a condition is coincident with the death of the fetus and evacuation of the uterus is necessary. In others abortion is the only recourse. As the liver very soon after this operation is performed becomes capable of again metabolizing the fats the prospects of the mother are fairly good. Professor Hendry informs me that, in his opinion, labor is more safely induced by bougies than by other methods in these severely emaciated, dehydrated patients. In the meantime, however, the physician may have considerable difficulty controlling the patient's acidosis on account of failure to retain an amount of carbohydrate sufficient to prevent insulin hypoglycemia. Rectal injections of 5 per cent glucose solution either in small repeated doses or by the Murphy drip method are useful. A solution of 5 per cent soda bicarbonate may also be administered in the same way. Normal saline may be injected subcutaneously to replace fluid lost, or, if necessary, saline or glucose may be given intravenously. The usual precautions in treating severe acidosis or coma should be carried out.

Insulin in infections

In our experience the incidence of infections in patients suffering from severe diabetes has decreased when insulin is employed in their treatment, and when infections occur in these patients healing proceeds as in non-diabetic patients. Joslin's (81) experience has been similar. A comparison of diabetics treated in the medical and surgical wards of a hospital shows much more favorable results in the former, due to the greater attention paid to proper dietetic treatment. The patient with a surgical infection is a much greater burden to carry from the medical standpoint than any other type of diabetic, even including the coma cases. In many cases that are badly infected or are in such a state of acidosis that surgical operations cannot be undertaken, it has been found that the institution of suitable dietetic treatment with insulin has resulted either in healing of the infection or in such a marked improvement in the general condition that any necessary surgical interference could be undertaken with relative safety.

Infection undoubtedly increases acidosis and reduces the tolerance. Pus, wherever found, should have free drainage at the earliest possible moment. In preparing a patient for immediate operation it is advisable to neglect glycosuria and concentrate attention on the acidosis for the time. The patient with glycogen in his liver is a much better risk from the standpoint of acidosis and, for this reason, he should be given sugar or other easily available carbohydrate along with insulin. Though diabetics with infection stand enormous quantities of insulin without bad results, it is preferable to use it in the proportion of one unit for each gram of sugar given. If the condition constitute a surgical emergency there should be no delay beyond that necessary for the ingestion of the soluble carbohydrate and the injection of the insulin. The anesthetics of choice are novocaine for intrathecal anesthesia, or paravertebral nerve blocking in suitable cases, or nitrous oxide oxygen by inhalation. Ether should be used sparingly, if at all; chloroform never. Local infiltration may sometimes be used but with caution, as gross damage to tissues is especially to be avoided in patients suffering from diabetes. In many less acute cases surgical interference does not demand other than the routine dietetic treatment. Simple drainage operations under nitrous oxide oxygen anesthesia may be undertaken with comparative safety. The patient, after operation, should be treated as a case of acidosis and gradually allowed suitable foods with enough insulin to abolish glycosuria and reduce hyperglycemia.

Diabetic patients suffering from pneumonia, tonsillitis, bronchitis, etc., should be given 400 to 600 calories more than the basal caloric requirement, with somewhat higher percentage of carbohydrate and protein than would ordinarily be used in treating the diabetic condition. The meals should be small, as attractive as possible and more frequently served. Foods high in carbohydrate content are used with advantage. Many investigators find it difficult to keep such patients aglycosuric because of the wide variations in tolerance exhibited from day to day. Strouse and Schultz (100) found in a series of five cases with various infections that a slight glycosuria persisted. We have previously noted the markedly increased tolerance to insulin during infections and suggest the use of larger doses of insulin in the treatment thereof. While admittedly more difficult

to accomplish in these cases, it is believed that the greatest care should be taken to keep the blood sugar as near the normal as possible.

The use of insulin in tuberculous diabetics has been discussed by several observers (101, 102). The usual effects of the drug are manifested in this disease. Its use, however, permits the administration of a larger caloric intake, so that the increased body nutrition assists in combating the spread of the pulmonary lesion. Insulin probably has no specific effect beyond this. Sansum believes that the tuberculous diabetic treated with insulin has the same opportunity for recovery as the non-diabetic patient suffering from this disease. Among our cases no cures have been observed as yet but several patients show astonishing degrees of recovery.

RULES FOR DIABETICS USING INSULIN

The dietetic treatment of diabetes does not come within the scope of this review, which is confined to the effects of insulin. There are, however, certain aspects of this part of the subject that require attention here. The results of various methods of treatment have a profound influence on the insulin requirement of any patient and also on its efficiency. Sufficient insulin will make any dietetic treatment work for a time. It cannot be too strongly insisted upon that each patient is worthy of the most careful study as an individual case and that he requires careful instruction and training in his own limitations: dietetic requirements, the principles of diabetic dietetics, the properties of insulin and how to administer it, the necessary experience in uranalysis and the symptoms of hypoglycemia and its treatment. No physician should undertake the treatment of considerable numbers of patients without submitting them to a preliminary course of training in a carefully regulated institution.

A memorandum accompanies each bottle of insulin, calling attention to the symptoms of hypoglycemia, its danger and its cure, and also to the method of its administration. Patients should also receive a memorandum in which the essential points in treatment are reviewed. For their convenience the following notes attached to the diet lists are furnished to our patients. Joslin makes use of a similar method for reminding the patient of his necessary limitations.

Information for patients

Weigh all food carefully. Learn to estimate quantity of food accurately in terms of protein, fat and carbohydrate content. Avoid monotony of diet by frequent change in the menu.

Arrange to get adequate rest; over-fatigue and emotional excitement must be avoided.

Careful regulation of the bowels, bathing, cleansing of the teeth, and other hygienic measures should be carried out.

Test the twenty-four hours' urine regularly for sugar and diacetic acid. If sugar appears in the urine reduce diet by one-third. After sugar disappears resume working diet gradually. The appearance of diacetic acid in the urine should be reported to your physician immediately.

Most so-called diabetic foods are dangerous. Refer to your physician before using any such article. Keep a note-book; record your progress and all questions you desire your physician to answer.

Complications, even slight ones such as colds, sore throats, boils, etc., are always serious in the diabetic. If ill from any cause notify your physician; go to bed; keep warm; reduce your diet by one-third; take $\frac{1}{2}$ pint of fluid per hour; take an enema. Coma can be avoided by prompt treatment.

The diabetic should always keep the body weight slightly below the normal.

Read the directions accompanying each bottle of insulin. Be sure your insulin is the proper strength.

Accurate adjustment of diet and insulin dosage is essential for successful treatment. Do not depart from the diet or dose of insulin found suitable unless your physician so advises. Avoid excessive exercise or heavy work.

Hypoglycemia, a condition caused by an overdose of insulin, is serious. Early symptoms are sudden hunger, weakness, fatigue, nervousness, pallor and flushing. The juice of one orange is usually sufficient to control the condition. More severe symptoms are sweating and tremulousness, marked anxiety, apprehension, delirium, unconsciousness or collapse. Lie down and rest. Notify your physician. Take a tablespoonful of corn syrup every ten minutes until

relieved. Always have some carbohydrate, such as oranges, corn syrup, candies, or sugar, readily available for such an emergency.

If vomiting or diarrhea occur or a meal is omitted from any cause discontinue insulin. Notify your physician. Do not resume insulin until your physician so advises or you are again able to eat your regular meals.

If your supply of insulin is exhausted decrease the diet by one-third and rest. Resume former diet when fresh insulin arrives.

Remember to sterilize your syringe as instructed before withdrawing the insulin from the bottle and inject the exact amount of solution required into the loose subcutaneous tissue of the body after carefully cleansing the skin with alcohol, ether or tincture of iodine.

Order insulin sufficient for one month. It will not deteriorate. Be sure to order your supply allowing plenty of time for delay in mails.

CONCLUSION

Since the discovery of insulin very great advances have been made in our knowledge of diabetes. The prognosis of the well-treated diabetic today as compared with his prognosis two years ago is very much improved. It should be remembered, however, that insulin is not a substitute for dietetic treatment of diabetes. On the contrary, the patient receiving insulin treatment is required to observe the limitations of his disease and of his diet more closely than a patient treated by dietetic means alone, and it is, therefore, not to be employed unnecessarily. However, should a patient respond favorably to dietetic treatment the very existence of insulin has a profound psychic influence upon him. He is aware that there is now a barrier between himself and death in coma if, for any reason, his tolerance should fail, infections occur, or any surgical emergency arise. Secure in this knowledge that he has to reckon with but little more than the usual hazards of life and death, his viewpoint on life has entirely changed. He no longer is depressed; he need no longer spend his days carefully tending the vital spark, but is enabled to enjoy life and work in a more normal manner than heretofore.

When properly employed, insulin replaces the lost hormone of the diabetic pancreas, restoring the power to metabolize sufficient food,

and makes it possible to treat patients in whom dietetic measures have failed. Despite the attendant disadvantages and inconveniences, insulin means life itself to these unfortunate patients hitherto condemned to a short precarious existence haunted by the imminent presence of death. The mental relief obtained is not less striking than the restored ability to work and to play now possible under insulin treatment.

For the treatment of the patient in acidosis or in coma, notwithstanding the benefits derived from other adjuvants in the treatment of the former and the occasional success of these methods in the latter, insulin has already become the most efficient weapon in the hands of those skilled in its use. Carefully controlled it must be; life-saving it undoubtedly is.

Physicians charged with the prolonged care of patients suffering from diabetes, rather than the treatment of the disease itself, have long since realized the difference between the diabetic to-day and the same patient two years ago. They recognize that the extra initial effort in treating and training patients is more than amply compensated by the increased health and strength of the patients and the simplicity of their subsequent guidance.

Our present knowledge of insulin only reveals more clearly further problems to be solved. Interesting in its history, fundamental in the recent advances in the physiology of metabolism, and of primary importance in the treatment of severe diabetes mellitus, many possibilities of insulin still lie in the future.

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DYSPNOEA

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I. INTRODUCTION

Shortness of breath, or dyspnoea, is not only a very frequent symptom, but likewise one with a wide variety of causes. In the pages which follow an attempt will be made to enumerate, analyze and evaluate the various factors which may play a part in its production.

To start with we must get clearly in mind just what is meant by dyspnoea. Etymologically the word signifies difficult breathing, and for the purposes of the present discussion let us assume that the difficulty is primarily subjective, in other words, that dyspnoea is in fact essentially a symptom, not a sign; the symptom that arises whenever in carrying on the respiratory function difficulty is encountered.

The respiratory movements, although they may be for short periods controlled by the will, are in the main involuntary and automatic.

The function of breathing goes on ordinarily without intruding itself into the field of consciousness. When, however, the respiratory organs do meet with embarrassment in the performance of their task, then not only do their movements enter the field of consciousness, but they enter it unpleasantly and produce discomfort, that is to say dyspnoea.¹

Regarding the production of dyspnoea, therefore, I think we may make the general statement that *dyspnoea will occur whenever the respiratory mechanism cannot with ease functionate to the extent that bodily processes require.*

Such a conception of the nature of dyspnoea simplifies our problem of analysis into a scrutiny, first, of those factors which determine the magnitude of the task which the respiratory mechanism must do, and second, of those which determine the ability of that mechanism to do it. That is to say, we must identify and discuss those factors which create a demand for pulmonary ventilation, and those which affect the efficiency of the bellows in meeting this demand. It becomes a question, if I may say so, of demand for and supply of pulmonary ventilation.

The prime determinant of the magnitude of the demand obviously is the rate of tissue respiration, or metabolism. The reason that we make respiratory movements at all is so that our cells may be supplied with the oxygen they require and be relieved of the carbon dioxide they produce.

Krogh has pointed out that in the lower organisms, those composed of a single, or of a few cells, and living in a fluid medium, the gaseous exchange between cells and environment takes place by the simple diffusion of the gases through the fluid medium and the cell membrane and protoplasm. As the animal organism becomes more complex such a simple arrangement will no longer serve. Diffusion is fast enough for very minute distances only. As the animal increases in size, therefore, diffusion has to be aided and a circulation becomes evolved. Here a moving fluid medium, by its act of motion, carries the respiratory gases from one part of the organism to another faster than they could go by diffusion alone. Such a system, how-

¹ Meakins has given the following excellent definition, "Dyspnoea is the consciousness of the necessity for increased respiratory effort."

ever, requires a pump to maintain circulation and stations at which the gases are exchanged first with the cell (the tissue capillary), and secondly with the outside environment, that is the respiratory organ in the usual sense. In the last the circulating medium or blood must have free opportunity for gas exchange with the outside environment be it water or be it air.

In the respiratory organs of the smaller air breathing animals, Krogh has further pointed out that diffusion alone again takes care of gas exchange. The smaller insects, for example, have a system of tracheae along which the gases diffuse. So short are these tubes that diffusion of itself carries the gases as fast as is necessary. No respiratory movements are required but as animals become larger diffusion again becomes inadequate and respiratory movements become necessary. So far has the respiratory organ itself become removed from the outside atmosphere that the latter must be actively pumped in and out, if the respiratory gases are to be exchanged with the blood at an adequate rate. The making of respiratory movements, therefore, is a price which the larger air breathing animal must pay in return for its greater size.

It is clear then that our search for the causes of difficulties in the way of external respiration, and so of dyspnoea, must start with tissue metabolism or internal respiration. We must, in other words, deal with the entire mechanism by which, in the higher animal, the tissue cells are kept, with respect to gases, in an environment best suited to their normal life. Such a mechanism will consist of a series of closely interrelated parts which for convenience we may outline very simply as follows: (1) *Tissue cells* which exchange gases by diffusion with (2) *tissue fluids* which in turn exchange gases by diffusion through the walls of the (3) *tissue capillaries* into the circulating fluid medium, that is the (4) *blood* which is pumped laden with its gases by the force pump, the (5) *heart* to the (6) *pulmonary capillaries* where again by diffusion² the gases are exchanged with the (7) *pulmonary air* which in turn is refreshed by the bellows or (8) *thorax* which continually washes it with the air of the (9) *outside atmosphere*.

² Certain physiologists, notably J. S. Haldane, believe that the lung epithelium may actively secrete oxygen from pulmonary air to blood, at least at certain times.

From this outline it becomes clear that the task imposed primarily by the metabolism of the tissue cells upon the external respiratory mechanism, or bellows, may be modified by, and will be dependent upon every link in the chain. We must therefore, in considering dyspnoea as we meet it clinically, bear constantly in mind the complex interdependence of the various elements in the great circulatory-respiratory mechanism, and recognize that abnormalities of any of these may provide the cause (of dyspnoea) for which we search.

II. THE METABOLIC DEMAND FOR PULMONARY VENTILATION

The subject of the rate of combustion within the tissues, that is to say of metabolism, provides material in itself for many monographs. For the purposes of the present discussion of dyspnoea, it will be necessary merely to state the fundamental laws governing metabolism and the several factors which may alter it in health or in disease.

In the first place, it may be said that the rate of metabolism is determined neither by the supply of fuel nor of oxygen. In the warm blooded animal it is so regulated that it never, except in disease, goes below a certain minimal or basal level. Of course metabolic rate is the direct expression of cellular activity, but in the warm blooded animal it appears that there is a basic level below which this activity never drops. Rubner has shown that the basal metabolism is chiefly related in warm blooded animals to the area of the body surface, and that expressed in terms of area, the basal rates for different members of a species, and even for different species, are remarkably constant.

For the normal man of twenty-five years the basal metabolism would be in the neighborhood³ of 40 calories per square meter of area per hour, and for a woman of the same age 37 calories. This in terms of oxygen and carbon dioxide would be in the neighborhood³ of 140 cc. of oxygen per square meter per minute and 112 cc. of carbon dioxide for the man, and 128 cc. of oxygen and 102 cc. of carbon dioxide for the woman. A man weighing 70 kgm. and 160 cm.

³ The exact amounts would depend on the Respiratory Quotient which denotes the kind of food being burned.

tall would have an area of 1.73 square meters, and a woman weighing 50 kgm. and 140 cm. tall, an area of 1.36 square meters. Such a man's gas exchange would be about 242 cc. of oxygen per minute and 194 cc. of carbon dioxide, and such a woman's 174 cc. of oxygen and 139 cc. of carbon dioxide.

Several things may increase cell activity, and therefore metabolic rate, over the basal value. These we must note briefly, especially to observe the relative magnitude of their effects. Chief among them is the performance of muscular work which has a far greater effect on the rate of combustion than any other factor. It is for this reason

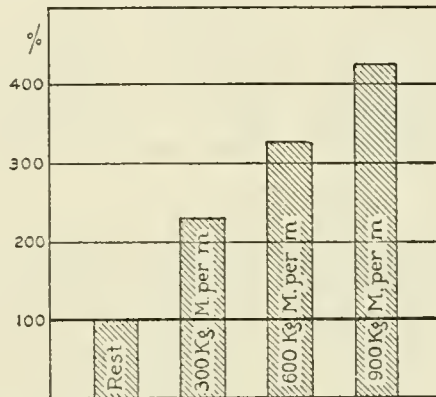


FIG. 1. METABOLISM DURING MUSCULAR WORK

Note the magnitude of the increments with varying amounts of work. This diagram shows the results obtained with the author as subject, the work being done on a stationary bicycle with ergometer attachment. Resting metabolism taken as 100 per cent.

that most dyspnoeas are, at least in their beginning, worse during exertion. The degree to which muscular work can increase metabolism is shown graphically in figure 1. It will be seen that during moderately hard work (900 kgm. m. per minute) the metabolism was increased more than fourfold. It is also interesting to note in figure 19, which shows the data of the same experiment in somewhat different form, that the pulmonary ventilation was increased in essentially the same proportion.

The effect of food which is perhaps the next most important accelerator of metabolism is slight when compared with that of work.

It is greatest in the case of the proteins, and in figure 2 is shown the magnitude of the rise after a large protein meal.

The secretion of certain of the endocrine glands, especially the adrenal and the thyroid, have a profound effect on metabolic rate. Both tend to elevate it, but the former does so rapidly (within an hour) while the latter does so slowly (a matter of several days).

Of other physiologic accelerators external cold may be mentioned. As outside temperature falls the warm blooded animal preserves his internal temperature first by diminishing the rate of heat loss. This

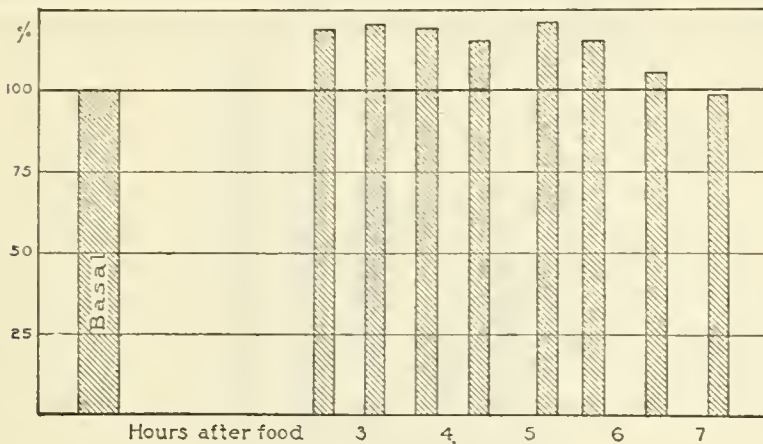


FIG. 2. METABOLISM AFTER MEAT INGESTION

The experiment was performed upon Dr. Aub. The amount of meat eaten was 500 grams. Compare the magnitude of the elevation after food with that during work shown in figure 1. The fasting (basal) metabolism is taken as 100 per cent.

method becomes insufficient after a time, and in order to prevent body temperature from falling, heat production has to be increased.

It will be seen, therefore, that the metabolism of normal persons will be at a basal level only when all those factors which may increase metabolism are absent: that is the individual must be at complete muscular rest, without food, in a surrounding temperature that is not chilly, and in a state of mental calm, the last named because of the effect of emotional stress upon the liberation of adrenalin. The subject who is in a state of excitement, even though making no muscular motion, may have an elevated metabolic rate.

The actual total daily metabolism of any normal person will consist first, of his 24 hour basal calories which, as we have seen, are closely proportional to his surface area;⁴ and secondly, of all the calories that are added for any of the causes mentioned, that is the taking of food, emotions, but chiefly those due to muscular work.

Passing now to disease we find several conditions in which the basal rate is abnormal. Most conspicuous among these are those diseases of the thyroid gland in which there is supposed to be an alteration in the gland's rate of secretion. In the clinical states known as hyperthyroid or thyrotoxic, such as exophthalmic goiter and toxic adenomatous goiter, there is a marked elevation in basal metabolic

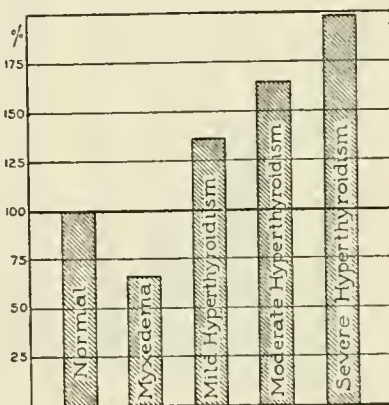


FIG. 3. THE BASAL METABOLISM OF A MAN WHILE IN A NORMAL STATE TAKEN AS 100 PER CENT, COMPARED WITH WHAT IT MIGHT BECOME IF HE DEVELOPED MYXOEDEMA ON THE ONE HAND OR MILD, MODERATE OR SEVERE HYPERTHYROIDISM ON THE OTHER

Compare with the physiologic changes shown in figures 1 and 2

rate, while in those known as hypothyroid, such as myxoedema and cretinism, there is a marked reduction. The magnitude of these changes is indicated in figure 3. They should be compared with the physiologic alterations shown in figures 1 and 2. It will be noted that in exophthalmic goiter there may be an increase of anywhere from 20 to 100 per cent and in myxoedema a similar decrease.

⁴ There are slight differences between the sexes and at different ages.

There are a few other diseases showing characteristic alterations in metabolic rate, especially the two forms of leukemia in which there are elevations of about the same magnitude as in exophthalmic goiter. In pernicious anemia there is occasionally a less marked elevation, and sometimes, and this is of especial interest with relation to dyspnoea, there may be a moderate rise in decompensated heart disease.

A reduction in metabolic rate is found in starving persons. This is not infrequently seen clinically in the starved or partially starved diabetic.

The changes found in fevers of all sorts are reasonably constant so DuBois has found. The human organism, he believes, follows to a certain extent a law like that of Van't Hoff for the speed of chemical reactions. As temperature rises, heat production rises. DuBois believes that the magnitude of the rise is in the neighborhood of 7.2 per cent for each degree Fahrenheit of fever. Thus a pneumonia patient with a temperature of 105°F. might be expected to have a metabolic rate increase of 47 per cent from his fever alone.

In concluding this brief résumé of the phase of the problem that has to do with the metabolic demand for ventilation, we may lay down as a general principle that, other things being equal, pulmonary ventilation will be proportional to metabolic rate. Substantially this principle has been formulated by Haggard and Henderson and called the *first law of breathing*. It will require some qualification as we proceed, and of course oftentimes other things are not equal. It is these other things that we are about to investigate, in doing so, however, let us keep constantly in mind the magnitude of the ventilatory task as it is primarily determined by metabolic demand.

III. GAS TRANSPORT BY THE BLOOD

As a first step in our study of how the metabolic demand becomes translated into actual ventilatory performance, it will be desirable to consider briefly the transport of the respiratory gases in the blood. This again is a matter upon which many volumes have been, and are yet to be, written. It is a field in which, because of wide-spread and active investigation, theories are constantly changing, information constantly advancing. The work has gone on in many countries,

Pflüger in Germany being one of the pioneers. In Denmark, Bohr, Hasselbalch, and Krogh; in England, Barcroft, and Haldane, and in this country L. J. Henderson, Y. Henderson, and Van Slyke have all made important contributions.

It is one of the basic principles of physical chemistry that gases go into physical solution in liquids in direct proportion to their partial pressure or tension. Another principle is that in a mixture of gases the tension exerted by any one gas is proportional to the actual concentration of that gas. Thus in atmospheric air there is 20.9 per cent of oxygen and the pressure of an atmosphere at sea level is 760 mm. of mercury. The partial pressure or tension, then, of oxygen in atmospheric air is

$$\frac{20.9}{100} \times 760 = 159 \text{ (mm. of Hg)}$$

All gases are not equally soluble, oxygen is far less so than carbon dioxide, but still the principle holds that the amount going into solution is proportional to the tension. When distilled water is exposed to atmospheric air, oxygen, nitrogen and carbon dioxide (though of the latter there is but an infinitesimal amount in atmospheric air) will pass from the air into solution in the water until an equilibrium is reached in which the tension of each gas is the same in both atmosphere and water. This is the saturation point for each gas at its existing tension.

As far as the physical solution of gases is concerned, blood plasma behaves just as does distilled water, except that solubility is slightly less in it. Both carbon dioxide and oxygen go into physical solution in plasma, and the relative amounts of them so physically dissolved are in direct proportion to their tensions. The transport of gases by the blood, however, can by no means be explained on the basis of physical solution alone. Blood will not only take up far greater quantities of oxygen and carbon dioxide than can be explained on the basis of their solubility, but a study of the amounts of gas taken up at different tensions shows that the capacity of the blood for either gas does not vary in direct proportion to tension. The reason for this is that each gas not only goes into physical solution in the blood

plasma, but forms chemical unions with substances in the blood as well. In the case of oxygen it is with hemoglobin, in the case of carbon dioxide it is largely with sodium and other kations to form bicarbonate.

The laws governing the transport of gases by blood can best be studied by exposing samples of blood to various gas mixtures in tubes known as tonometers. In such tubes, by slow rotation at body temperature, a thin film of blood is exposed to the gas mixture and in

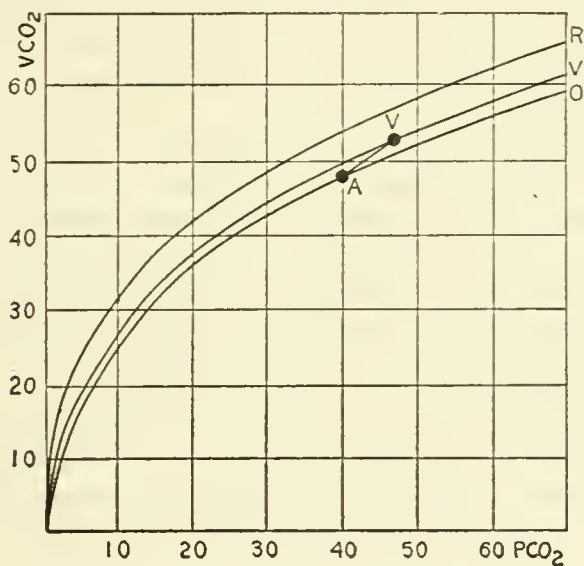


FIG. 4. THE CARBON DIOXIDE DISSOCIATION CURVE FOR BOCK'S BLOOD

The ordinate VCO_2 indicates the blood's content of carbon dioxide in volumes per cent; the abscissa PCO_2 the partial pressure of carbon dioxide in millimeters of mercury. The curve O is for completely oxygenated blood; the curve R for completely reduced blood; the curve V , lying between the two, is that of ordinary venous blood. A is the actual arterial point in vivo, and V the venous.

a short time tension equilibrium is established. This process is known as equilibration. Analysis, then, of the air in the tonometer gives the tension of gases in the air and in the blood as well for they are in equilibrium, and analysis of the blood gives the actual volume of gas taken up per volume of blood. By exposing blood to a variety of tensions in this way curves may be plotted which show the behavior

of blood in vitro for all combinations of the two gases. I am indebted to Bock, Field and Adair for the privilege of using a series of such curves determined very accurately during the last year for Bock's blood. Let us proceed to examine them.

In figure 4 we have the so-called carbon dioxide dissociation curve. In this plot the abscissa PCO_2 gives the tension of carbon dioxide in the equilibrating atmosphere (and hence in the blood also) as determined by air analysis, and the ordinate VCO_2 , the amount of carbon dioxide taken up by the blood in volumes per cent as determined by blood gas analysis. The curve *O* shows the behavior of Bock's blood for all tensions of carbon dioxide between 0 and 70 mm., in the presence of an excess of oxygen.

If now we plot a curve for blood similarly treated except that oxygen is excluded (nitrogen taking its place) we get the one shown at *R*. These two curves, similar in shape but at somewhat different levels, represent the combining power of Bock's blood with carbon dioxide, in the case of *O* when his hemoglobin is all oxygenated, that is to say, when it is all oxyhemoglobin, and the case of *R* when it is all reduced hemoglobin. We may call *O* his oxygenated carbon dioxide dissociation curve and *R* his reduced curve.

Venous blood contains less oxygen than arterial yet it is not completely reduced, the dissociation curve for venous blood therefore lies somewhere between curves *O* and *R*. Its actual position for the usual oxygen tension of Bock's venous blood is shown by curve *V*, (fig. 4).

These curves of course show the behavior of the blood under a far wider variety of conditions than ever occurs in the body. Exactly what is happening in the body may be discovered by obtaining (by arterial puncture) a specimen of arterial blood and determining its carbon dioxide content. The appropriate ordinate for this content is found and the point *A*, which is its intersection with the curve *O*. (This curve may be used to represent arterial blood for arterial blood may be considered, at least for the present, as fully oxygenated.) The position of the point *A*, therefore, shows both the content and tension of carbon dioxide in Bock's arterial blood. In a similar way the point *V* may be placed upon the curve *V* to represent the content and tension of his venous blood. A line connecting these two points,

as was first shown by Christiansen, Douglas and Haldane, represents what actually happens with respect to carbon dioxide in the blood as it exists in the body. It has recently been shown by L. J. Henderson to be a curve, not a perfectly straight line.

Let us pass from this to the matter of oxygen. We can equilibrate blood with various oxygen mixtures in exactly the same way as with carbon dioxide and plot dissociation curves. In figure 5 are shown those obtained with Bock's blood. Two curves are given, they represent his blood's behaviour with respect to oxygen but at different

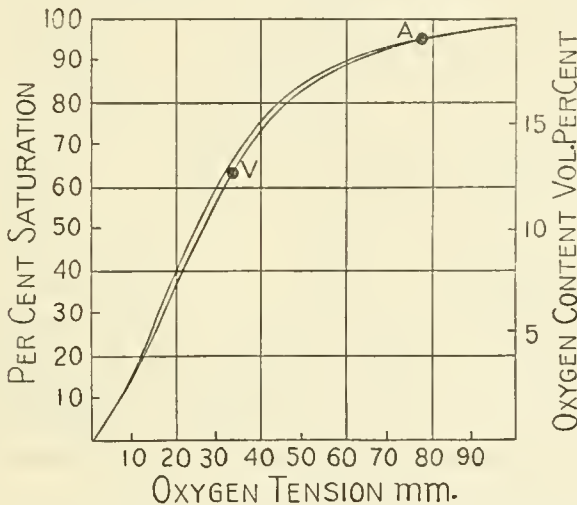


FIG. 5. THE OXYGEN DISSOCIATION CURVE FOR BOCK'S BLOOD

The ordinate denotes the quantity of oxygen contained in the blood, shown at the right as volumes per cent, and at the left as percentage saturation. The abscissa denotes the tension of oxygen in millimeters of mercury. The upper curve is that of arterial blood, the lower that of venous. *A* is the arterial point, and *V* the venous.

tensions of carbon dioxide from those of his venous and arterial blood. Just as changes in the tension of oxygen altered the level of the carbon dioxide dissociation curve, so do changes in carbon dioxide affect that of the oxygen dissociation curve. In figure 5 the abscissae represent oxygen tensions, and the ordinates the blood's oxygen content in volume per cent.

The actual carbon dioxide tensions of Bock's arterial and venous blood are 40.3 and 47 mm. respectively. The upper and lower curves represent the true levels of oxygen dissociation of his arterial and venous blood respectively. The oxygen *A* and *V* points are placed upon the *A* and *V* curves as was done in the case of the carbon dioxide points, and a line drawn between them again would represent what actually takes place in the body, this time with respect to oxygen.

Several characteristics of these curves should be emphasized. First their shapes; it will be observed that the carbon dioxide curve rises sharply at first and then more gradually, but nevertheless it continues to rise throughout its whole extent. The oxygen curve, on the other hand, eventually approaches a horizontal line. This in normal bloods is reached when they contain about 20 volumes per cent of oxygen; that is to say, 20 volumes per cent is the total oxygen capacity regardless of tension. For this reason, with respect to oxygen, we speak of percentage saturation. When we say a blood is 100 per cent saturated with oxygen we mean that it has taken up in chemical combination all of which it is capable; when 50 per cent saturated, one half of which it is capable, etc. In figure 5 the percentage saturation of Bock's blood is shown by the left hand column of figures, the actual content at the right. The dissociation curve shows, at any given oxygen tension, to what extent the total oxygen capacity of the blood is used, or in other words, the relative amounts of oxy- and reduced hemoglobin present.

We cannot speak of the percentage saturation of the blood with carbon dioxide as we can with oxygen, because of the fact that its curve does continue to rise throughout its extent. Hemoglobin gets fully oxygenated at 100 mm. of oxygen tension or less and increasing the tension beyond that will not make it take up any more. Of course there is a small increase in dissolved oxygen as pressure rises, but the solubility of oxygen is so slight that this may be disregarded. With carbon dioxide, on the other hand, no such level is reached and the amount not only in physical solution, but chemically combined as well, goes on increasing as carbon dioxide tension increases, way beyond any level that is ever reached in vivo.

These relationships can perhaps be more readily understood by reference to figure 6 in which the two curves (oxygen and carbon

dioxide) are shown together. Not only does the difference in shape become more apparent, but likewise the difference in level. Both of these curves show a very beautiful adaptation on the part of the organism to its environment. Thus it will be seen in the case of the oxygen curve that at a tension of 80 mm. or over, the blood will be 95 per cent or more saturated. In the lungs the area over which the blood is exposed to the air in the alveoli is enormous, about 90 square

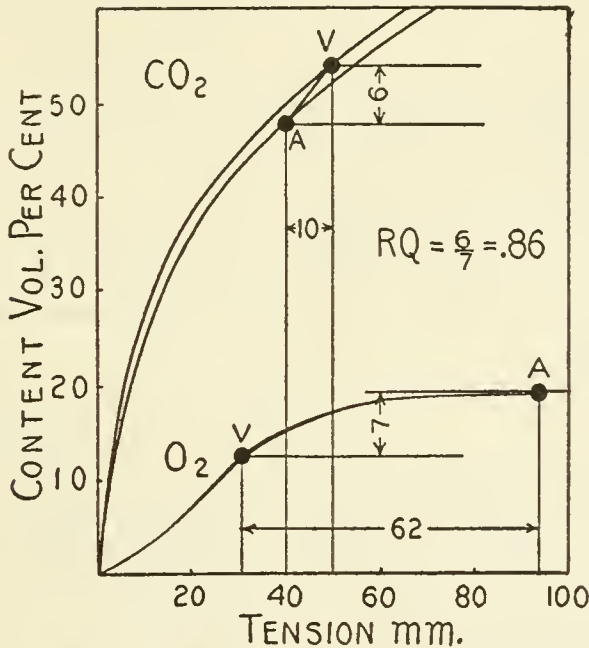


FIG. 6. THE OXYGEN AND CARBON DIOXIDE DISSOCIATION CURVES SHOWN TOGETHER

The ordinate represents the blood content of either gas; the abscissa, the tension. Attention is called to the comparatively short arc of the carbon dioxide curve which is used in vivo, and the comparatively long one of the oxygen curve. It will be noted that the tension difference between arterial and venous blood is 62 mm. in the case of oxygen, and only 10 mm. in the case of carbon dioxide. Nevertheless, such are the shapes of the curves that, in this particular instance, there are transported 6 volumes in the case of carbon dioxide and 7 in the case of oxygen, giving a respiratory quotient (R. Q.) of 0.86.

meters as calculated by Zuntz. The blood, therefore, rapidly comes into equilibrium with the air. In a normal person the air of the alveoli contains in the neighborhood of 14 per cent of oxygen which would have

a tension of about 100 mm., this means that in the lungs the blood, if the subject is breathing atmospheric air, will always readily become nearly completely saturated with oxygen. Conversely, in the tissues where oxygen tension is low, the blood will easily liberate oxygen. In the lungs then conditions are such that a full load of oxygen is readily taken in, and in the tissues such that unloading is easy.

The same in general is true with respect to carbon dioxide. We should carefully note, however, that in the case of the oxygen curve a long arc is used *in vivo* which means wide fluctuations in oxygen tension between arterial and venous blood (in the case shown in figure 6 it is 62 mm.), while with the carbon dioxide curve, on the other hand, a comparatively short arc is used, which means but slight difference in tension between arterial and venous blood (in figure 6 it is 10 mm.) Nevertheless, such are the shapes of the curves that the volumes of gas transported, as can again be seen in figure 6, are not very dissimilar, the actual relation between them which is the so-called respiratory quotient or R.Q. being dependent on metabolism.⁵

The shifts in the curves caused by the opposite gas are of importance; in figure 5 it may be observed that the greater the carbon dioxide tension, the less oxygen capacity, and vice versa; and in figure 4, that with the carbon dioxide curve the greater the oxygen tension, the less the carbon dioxide capacity (at a given CO_2 tension). This shift in the oxygen curve was first demonstrated by Bohr, Hasselbalch and Krogh, and later studied more extensively by Barcroft, while that of the carbon dioxide curve was first shown by Christiansen, Douglas and Haldane.

The importance of these interrelationships has been thoroughly analyzed by L. J. Henderson; in brief it is this: in the lungs the fact that the blood takes up oxygen makes it easier for it to give off carbon dioxide and vice versa, and in the tissues the fact that oxygen is unloaded makes the taking up of carbon dioxide easier and vice versa. In other words, the taking up of one gas always facilitates the discharge of the other. They have a mutually unloading effect, which must be regarded as a very beautiful adjustment indeed.

⁵ Unless altered by over or under breathing, as will be discussed later.

With respect to its respiratory function I think the proper concept to hold of the blood is that of a substance peculiarly well adapted, as we have seen, to transport the respiratory gases, bounded on the one hand by the tissues with which it exchanges the gases, and on the other by the air in the lung alveoli (alveolar air) with which it also exchanges the gases. The exchange in each case is probably due to simple physical diffusion⁶ and therefore is dependent upon the tension difference for each gas between blood and tissue or between blood and alveolar air.

The tension differences in the tissues depend upon metabolism, circulation rate, and ventilation rate; those in the lungs upon exactly the same; the last, because as can easily be seen, the composition of the alveolar air will be determined by the rate at which gases are added to or subtracted from it by exchange with the blood, and by the rate at which it is washed or diluted with outside air, that is to say, by the rate of pulmonary ventilation.

In other words, in the great function of respiration in the higher animals, we have the triad, metabolism, blood flow, and pulmonary ventilation all indissolubly linked, the two last adjusted to meet the requirements of the first.

IV. BLOOD BICARBONATE IN RELATION TO PULMONARY VENTILATION

We are now in a position to consider the manner in which the second and third members of our triad are adjusted to meet the requirements of the first.

With respect to carbon dioxide, it may be readily seen, that the pulmonary ventilation and blood flow must both be so regulated as to eliminate the gas in the lungs at the same rate that it is produced in the tissues, and with respect to oxygen they must likewise be so regulated as to lead to the absorption of the gas in the lungs at the same rate that it is called for by the tissues. A tension difference between blood and alveolar air must be created for each gas sufficiently great, at their respective rates of diffusion, to allow the required oxygen to enter the blood and the carbon dioxide produced to leave it. These

⁶ Haldane's school believe that under certain circumstances the lung alveolar membrane actively secretes oxygen inward, this view, however has not been generally accepted.

tension differences we have already seen are dependent, with a constant rate of metabolism, upon both circulation rate and ventilation rate.

The key to the problem of how they are adjusted was found by Haldane and Priestley in 1905 and consisted in their discovery of the following facts:

1. That while the oxygen tension of the alveolar air in normal persons may vary widely, that of carbon dioxide varies very little, even in the face of marked variations in total barometric pressure or in rate of metabolism.

2. That the addition of carbon dioxide to the inspired air even in very small amounts causes significant increments in pulmonary ventilation, but

3. That relatively wide variations in the oxygen tension of the inspired air may occur with but slight changes in ventilation.

The movements of the external respiratory apparatus are known to be controlled by a center in the medulla, the so-called respiratory center. If this center is destroyed, no respiratory movements can occur. From their observations on alveolar air Haldane and Priestley concluded that this center had for its normal stimulus the carbon dioxide tension in the blood reaching it from the lungs. The blood, which is the arterial, is essentially in tension equilibrium with the air within the alveoli. Any increase in the carbon dioxide tension in the alveolar air, and hence in the blood reaching the center, they believed stimulated the center and accordingly increased pulmonary ventilation, while any decrease had the opposite effect.

In our study of the dissociation curves in the previous section, we noted that in Bock's blood the fluctuations in tension between arterial and venous blood, with respect to carbon dioxide, were slight compared with those of oxygen. We also noted that at a certain tension of oxygen blood becomes saturated with that gas, and that increasing the tension above that point has no effect except to drive a little more into physical solution which, because of the low solubility of the gas, is practically a negligible quantity; oxygen is twenty-six times less soluble than carbon dioxide. In the case of carbon dioxide this principle does not hold. With the latter gas the higher the tension, the greater will be the amount that the blood will take up.

These things being true it will be seen that a ventilation rate sufficiently great at any given rate of metabolism to create an alveolar oxygen tension of 90 mm. will saturate with oxygen the blood of a normal person, and that increasing it will not increase the rate of oxygen absorption. With carbon dioxide on the other hand, the greater the tension difference between blood and alveolar air, the faster will this gas come off. It will also be seen that if the ventilation is raised to such a point that the elimination of carbon dioxide in the lungs takes place at a faster rate than that at which it is produced in the tissues, its quantity and tension in the blood will be diminished. By excessive breathing, in other words, the body can be depleted of

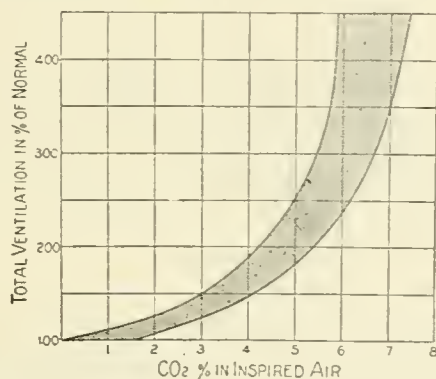


FIG. 7. THE VENTILATION REACTION TO INCREASING CONCENTRATIONS OF CARBON DIOXIDE IN THE INSPIRED AIR

The shaded zone includes the reaction curves of normal human subjects. The total ventilation with increasing concentrations of carbon dioxide in the inspired air is expressed in per cent of that when the inspired air contains no carbon dioxide. (After Peabody).

carbon dioxide, it cannot, however, by the same process be surfeited with oxygen. If the breathing is voluntarily increased so that carbon dioxide is washed out to such an extent that its tension in the blood falls below the threshold of the respiratory center, then cessation of respiration, or apnoea, will ensue and will continue until the metabolism has restored the lost carbon dioxide to the tension required by the center. This apnoea production by forced breathing was another of the discoveries of Haldane and Priestley and adds strong weight to their contention that carbon dioxide tension regulates lung ventilation.

The ventilation reaction to increased concentrations of carbon dioxide in the inspired air is another strong bit of evidence of the extreme sensitivity of the respiratory center toward the tension of this gas. The reaction is very constant in normal persons and its magnitude is shown in figure 7 taken from Peabody.

Ordinarily carbon dioxide plays the major rôle in regulating the respiration, and the rate of ventilation is adjusted to carbon dioxide requirements. For purposes of oxygen absorption alone the ventilation is ordinarily in excess. This can be shown by the findings in any normal person.

Let us take Bock for example. At rest his metabolism produces 200 cc. of carbon dioxide per minute and calls for 225 cc. of oxygen per minute. His oxygen dissociation curve shows that at 100 mm. of oxygen tension his blood becomes nearly fully saturated. To maintain the tension of oxygen in the alveolar air at that level when it is being absorbed at the rate of 225 cc. per minute would require a ventilation of the alveoli of 3.00 liters per minute. But at this rate and with a carbon dioxide output per minute of 200 cc., the alveolar carbon dioxide tension would be about 49 mm.; his actual alveolar carbon dioxide tension, however, as the curve shows, is nearer 40 mm., and to preserve that he requires a ventilation of his alveoli of 3.67 liters per minute. In other words, 3.00 liters per minute would saturate his blood with oxygen and give him the oxygen he requires but it takes 3.67 liters per minute to keep his carbon dioxide tension down to the level that his respiratory center demands.

Having observed the facts, we may now seek the explanation. That bodily economy permits wide fluctuations in oxygen tension, but only very slight ones in carbon dioxide tension, seems to point to the conclusion that a relatively fixed carbon dioxide tension serves some useful purpose. Fortunately data are available which show that this is true.

The cells of the warm blooded animals we know cannot live unless certain conditions of their immediate environment are kept from varying, save through a slight range; for example, the temperature, and more particularly the chemical reaction. It is well known that changes in blood reaction beyond a narrow limit are incompatible with

life, and the preservation of blood neutrality is as necessary to the life of the cells as is the oxygen their metabolism calls for.

The mechanism for stabilizing blood reaction is inseparably interwoven with that of gas exchange from its very origin, this because one of the respiratory gases, carbon dioxide, is at the same time in the form of carbonic acid H_2CO_3 , the chief source of acid in the blood. The amount of acid delivered to the blood then is going to vary directly with the metabolism, therefore the neutrality regulating mechanism, like the pulmonary ventilation, must be adjusted to metabolic rate. In fact we find that the pulmonary ventilation itself is one of the most important elements in the neutrality regulating mechanism.

Blood is a highly buffered solution and L. J. Henderson has shown that of the various equilibria upon which its reaction depends, that between carbonic acid and the bicarbonates is by far the most important, and in fact for all practical purposes, the hydrogen ion concentration of the blood may be said to depend upon it, thus,

$$(\text{H}^+) = K \frac{(\text{H}_2\text{CO}_3)}{(\text{BHCO}_3)}$$

in which (H^+) is the hydrogen ion concentration, (H_2CO_3) the concentration of carbonic acid in solution and (BHCO_3) the concentration of bicarbonates in the plasma. K is the reaction constant under the Mass Law, and since it is a constant we may neglect it.

A little scrutiny of this equation will give a very clear understanding of the interrelationship between gas exchange and neutrality regulation. Let us consider its terms separately. (H_2CO_3) the numerator of the fraction, which is the concentration of free carbonic acid in the blood, is of course going to depend upon the balance between the rate at which carbon dioxide is added to the blood and hence upon the metabolism, and the rate at which it is removed and hence also upon pulmonary ventilation. (BHCO_3) the denominator, which is the concentration of bicarbonate, is obviously going to depend upon the supply of basic radicals in the blood. (H^+) or the hydrogen ion concentration is therefore dependent on the ratio between free carbonic acid and base.

The problem of the body may be briefly stated thus: sufficient pulmonary ventilation must be supplied to remove the carbon dioxide produced by metabolism, but at the same time its rate of removal must be so adjusted to its rate of addition to the blood that its tension will

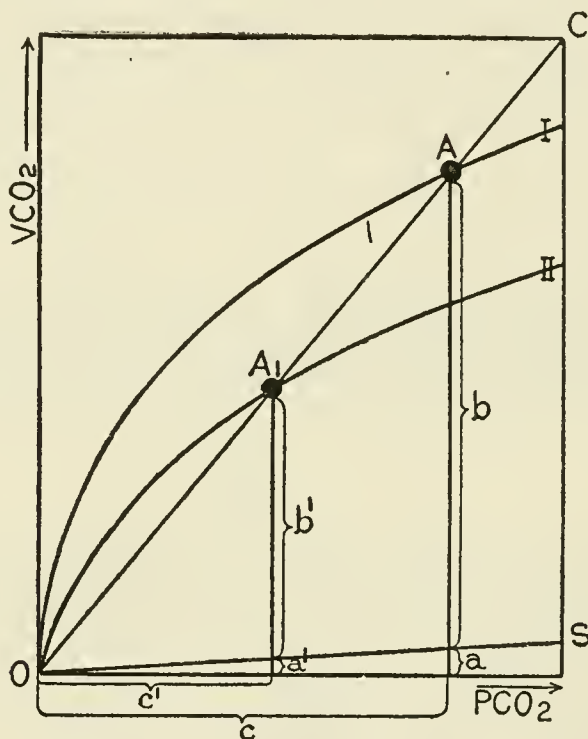


FIG. 8. THE CARBON DIOXIDE DIAGRAM OF THE BLOOD

The ordinate VCO_2 denotes the blood's content of carbon dioxide; the abscissa PCO_2 , the carbon dioxide tension. The diagonal OC denotes the hydrogen ion concentration of normal blood. The diagonal OS divides the ordinates for all points falling upon OC into the portion of carbon dioxide physically dissolved (a), and that chemically bound (b). I is a normal level carbon dioxide dissociation curve; II is a low level curve. A is the arterial point of I; A_1 is the arterial point of II.

bear a certain nearly fixed ratio to the blood bicarbonate, this in order that the blood reaction may be kept from more than slight alteration.

The problem can perhaps be made clearer by the use of the geo-

metric method of representation used by Haggard and Y. Henderson. Let us therefore return to the dissociation curve. In figure 8 two such curves are shown, one at a normal level (curve I), one at a low level (curve II). The ordinates of these curves, it will be remembered, are the total carbon dioxide content of the blood, this total being made up of dissolved carbon dioxide (H_2CO_3) and combined carbon dioxide (BHCO_3). The first of these, however, we have already noted, is directly proportional to the carbon dioxide tension, therefore Haggard and Henderson draw a line OS which represents the amount of carbon dioxide in physical solution for any tension of that gas. This line cuts the ordinates of our curves and the portion of each ordinate below OS represents the dissolved, while the portion above represents combined carbon dioxide. But these two portions of the ordinates are in fact identical with the two terms of the equation in one algebraic expression, and if we call the lower portion, a , and the upper, b , we have

$$\frac{(\text{H}_2\text{CO}_3)}{(\text{BHCO}_3)} = \frac{a}{b}$$

therefore $\frac{a}{b}$ is also equal to the hydrogen ion concentration. For curve I the particular ordinate drawn is the one which passes through the arterial point A . It will be seen that for no two points in that particular curve will $\frac{a}{b}$ have the same value. If, however, we pass to curve II, the arterial point of which A' lies on the same straight line OC as does A , then we find that $\frac{a}{b} = \frac{a'}{b'}$ because the triangles involved are similar. The line OC , therefore, denotes a certain hydrogen ion concentration, and other straight lines may be drawn through O representing other hydrogen ion concentrations, and our carbon dioxide dissociation curve (with its arterial point) or carbon dioxide diagram as Haggard and Henderson call it, shows us not only the blood carbon dioxide tension and content, but the concentration of dissolved and combined carbon dioxide and the hydrogen ion concentration as well.

For ordinary use we need not construct the line OS, for just as for the points A and A' is it true that

$$a:b = a':b'$$

so is it also true that

$$a + b:c = a' + b':c'$$

but $a+b$ equals the total blood carbon dioxide content while c equals carbon dioxide tension, so we can for every day use think of the carbon dioxide content as determined by blood gas analysis as equivalent to (BHCO_3) , and the carbon dioxide tension as determined by air analysis as equivalent to (H_2CO_3) , and their ratio, which is shown by the position of their intersection (the A or V points) when related to radii drawn through O , gives the (H^+) or blood reaction.

Carbon dioxide diagrams of this sort will answer the problem raised by the title of this section, namely what the blood bicarbonate level has to do with pulmonary ventilation.

As we study the blood in disease under certain circumstances we find carbon dioxide curves occupying positions lower than normal, as for example curve II in figure 8. If, however, we plot the arterial points for such curves, we often find as is shown in curve II, that there is little or no tendency toward change in hydrogen ion concentration. The arterial points tend to fall on a single diagonal or on diagonals in close proximity. This fact has led to the modification of the original Haldane-Priestley theory to the belief that what the respiratory center really reacts to and what in truth regulates lung ventilation is blood hydrogen ion concentration, not carbon dioxide tension. It is to preserve blood neutrality, not blood carbon dioxide tension, that the mechanism is designed. Of course with dissociation curves at a constant level carbon dioxide tension will remain constant as well as hydrogen ion concentration, but when the level is changed by disease or in other ways, as in curve II, it is found that it is the hydrogen ion concentration which the organism seeks to keep constant, not the carbon dioxide tension. This principle was first discussed by Henderson in 1909 and was later developed by Winterstein and Hasselbalch and accepted by Haldane.

Consider the points A and A' , they both fall on the same diagonal OC , and hence their hydrogen ion concentration is the same. Their

abscissae, however, show that in the case of A the carbon dioxide tension is much higher than in that of A'. The blood and alveolar air we have noted are in tension equilibrium, therefore in order to preserve a normal blood reaction, it is clear that the subject whose curve lies in the position II must ventilate his alveoli at a much higher rate than the subject whose curve lies in position I. Now the level of the curve is determined by the amount of base available to form bicarbonates. The curve level therefore, or blood bicarbonate, will determine the amount of pulmonary ventilation necessary to eliminate a given quantity of carbon dioxide per unit of time. In health the curve level varies but little, in disease it may vary widely. What factors may cause it to vary will be discussed later, the point that I should like to emphasize now is that to accomplish the elimination of a given quantity of carbon dioxide and at the same time to preserve normal blood reaction will require more lung ventilation for the person with a low level curve than for one with a higher one. With regard to the amount of work that the pulmonary bellows has to do, there is greater economy of effort with high than with low levels of blood alkali. Why the curves of normal men lie at just the level they do is undoubtedly due to various factors. For our present purposes it is interesting to note that their level is such as to secure a rate of ventilation ordinarily in excess of the requirements for oxygen absorption which may possibly be regarded as, in part at least, a safeguard against possible asphyxia.

We can conceive a curve lying at a considerably higher level than that of the normal one shown. An individual with such a curve would require less ventilation as far as CO₂ elimination is concerned, but if he had less ventilation he would have a lower alveolar oxygen tension, and if this process were carried far enough the point would come when alveolar oxygen tension would fall so low that the blood would no longer get saturated in the lungs and anoxemia would result. For example, a man having an oxygen absorption of 200 cc. per minute, at a ventilation of 3.3 liters would have an alveolar oxygen tension of about 110 mm., but if the height of his carbon dioxide curve were doubled as far as CO₂ elimination and acid-base balance is concerned, he could get along with half that or 1.6 liters. But at this rate of ventilation his alveolar oxygen tension would be but 55 mm. which

reference to figure 5 will show is not adequate to saturate his blood. He would be constantly in danger of oxygen want.

The sum and substance of the relation of gas exchange and neutrality regulation to pulmonary ventilation has been very lucidly stated by Haggard and Henderson in what they call first and second laws of breathing. The first we have already alluded to incompletely in an earlier section. They may both be now set forth in full.

The first is as follows: "At any one level of the CO_2 dissociation curve or arterial alkali, the pulmonary ventilation varies directly as the mass of CO_2 eliminated, and the alveolar and arterial tension of CO_2 therefore remains constant."

The second is the corollary of the first, and is this, "At different levels of the dissociation curve the pulmonary ventilation, per unit mass of CO_2 eliminated, varies inversely, and the arterial CO_2 tension, therefore, varies directly, as the amount of alkali which in the condition of the blood at the time, as expressed in its dissociation curve, will afford the normal C_H " (hydrogen ion concentration).

In somewhat simpler language the essential points of the first law amount to this,—*with a constant carbon dioxide tension ventilation will vary directly with carbon dioxide output*,—and those of the second law to this,—*with a constant carbon dioxide output ventilation will vary inversely as the carbon dioxide tension*. These two which follow of necessity from the carbon dioxide diagram will make clear many of the phenomena of respiration and neutrality regulation which without them are often difficult to comprehend. They of course do not hold when the blood reaction is shifted or the sensitivity of the respiratory center changes.

V. BLOOD FLOW IN RELATION TO PULMONARY VENTILATION

Of the rate and method of regulation of the blood flow far less is known than in the case of pulmonary ventilation. The latter can be readily measured by respiration apparatus; the former can only be estimated by indirect means.

So far two general principles have been employed to determine the rate of circulation in man. One, typified by the method of Krogh and Lindhard, depends upon the observation of the rate of

absorption in the lungs of a neutral gas such as nitrous oxide which forms no chemical union with blood but merely goes into physical solution. The other, the principle of Fick, depends upon the calculation of the minute volume of the blood stream from either of the respiratory gases by determining its rate of exchange and its concentration in the arterial and venous blood, thus:

$$\text{B.F.} = \frac{\text{O}_2}{(\text{aO}_2) - (\text{vO}_2)} \times 100 \text{ or } \text{B.F.} = \frac{\text{CO}_2}{(\text{vCO}_2) - (\text{aCO}_2)} \times 100$$

in which B.F. is blood flow in cubic centimeters per minute, O_2 oxygen absorption per minute, (aO_2) arterial oxygen content in volumes per cent, (vO_2) venous oxygen content in volumes per cent, and in which CO_2 is carbon dioxide elimination in cubic centimeters per minute, (vCO_2) venous and (aCO_2) arterial carbon dioxide content in volumes per cent.

Thus if a given subject's oxygen absorption were 200 cc. per minute, his arterial oxygen content 20 volumes per cent and his venous is volumes per cent, his blood flow would be 4000 cc. per minute or 4 liters.

Both principles are open to criticism, the Krogh-Lindhard because it is believed by some that the respiratory gymnastics through which the subject has to go in order that the required air samples be obtained disturbs his circulation to such an extent that the results are of little value. The Fick principle is criticized because of the extreme difficulty of obtaining accurate measurements upon the mixed venous blood. For the purposes of the present paper it is not necessary to enter the controversy concerning these two. It is, however, desirable to know that such a controversy exists and to appreciate in reviewing the available data that they are all open to question.

In table 1, I have collected from the literature and from personal work certain estimations of the blood flow in man. In this table are shown the subject's blood flow in liters per minute, the pulse rate, and the output of the heart per systole, which last is of course the total flow divided by the pulse. There is also a column labelled coefficient. This factor is an interesting one and needs a word of comment. We noted with regard to ventilation that the rate was

apparently in excess of what would be needed for purposes of oxygen absorption alone. So too in the case of blood flow there is what appears to be at first sight a rate in excess of what might theoretically be required simply to transport oxygen.

The blood, we know, has a certain capacity for oxygen, this being incidentally proportional to its hemoglobin content. A person with 100 per cent hemoglobin according to the ordinary scale has an oxygen capacity of approximately 20 volumes per cent, one with 50 per cent hemoglobin just half of that. The blood as it passes through the lungs has an opportunity to become almost completely saturated (about 95 per cent in point of fact). The venous blood returning to the lungs still has a considerable load of oxygen, perhaps 15 volumes per cent, which would mean that it was seventy-five per cent saturated. In other words, of the total capacity of the blood to carry oxygen there is actually used in this instance only 95 to 75, or 20 per cent. This proportion of the oxygen carrying power actually in use has been called by Krogh and Lindhard the "coefficient of utilization of the oxygen carrying capacity." It is an interesting factor for it is obvious that with regard to oxygen transport it is an index of the relative economy of the circulation.

For example, in figure 9, I have shown quite diagrammatically two arrangements. Let us suppose a normal person with 100 per cent of hemoglobin and an oxygen capacity therefore of 200 cc. per liter, and a metabolic call for oxygen amounting to 200 cc. per minute. If now with respect to oxygen transport such a subject used his circulation to its greatest possible efficiency, he would in his tissues remove all the oxygen and return venous blood completely reduced. Under these circumstances it is evident that he would require a blood flow of but one liter a minute, and would have a coefficient of 100 per cent. (To simplify the present illustration I am supposing that the blood becomes 100 per cent saturated in the lungs; as stated before, actually it becomes about 95 per cent saturated). This arrangement is shown in the upper part of the diagram. It would never occur in health but might be approached in disease. What happens in health is much more like the arrangement shown in the lower part of the diagram where four liters of blood flow per minute and where the venous blood returns 75 per cent saturated,

giving a coefficient of 25 per cent. Under the latter circumstances it requires four times as much work on the part of the heart to accomplish the same result; but we shall see later why for other reasons this higher flow may be desirable.

Table 1 shows that the different methods give results widely apart in some instances for the blood flow, the coefficient and the systolic output during rest; the main stumbling block is the value of the coefficient. Of course we can draw blood from an arm vein and determine its oxygen content, but the blood in the arm vein may be different from that in the mixed venous blood that reaches the heart. The

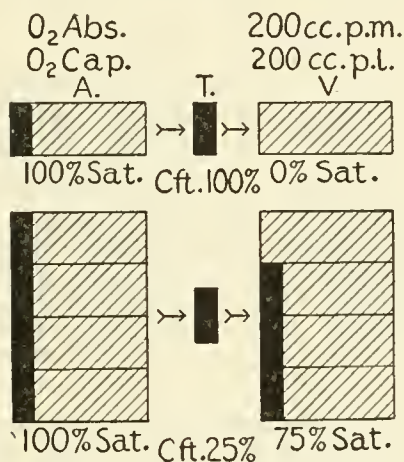


FIG. 9. SCHEMA OF THE BLOOD FLOW IN A NORMAL PERSON

The large rectangles denote liters of blood; the black portions denote volumes of oxygen. It is assumed that the individual's oxygen absorption is 200 cc. per minute, and that the oxygen capacity of his blood is 200 cc. per liter. Two theoretical alternative conditions of the blood flow are shown. In the upper, one liter of blood circulates per minute. In the lungs (*A.*) it takes up 200 cc. of oxygen. This entire load is left in the tissues (*T.*). The blood on returning to the lungs (*V.*) therefore contains no oxygen. The coefficient of utilization of oxygen carrying power (*Cft.*) is therefore 100 per cent. In the lower, the blood flow is 4 liters per minute. Four liters of blood take up 800 cc. of oxygen in the lungs, but since again only 200 cc. are unloaded in the tissues, the blood returning to the lungs contains 600 cc., that is to say, is still 75 per cent saturated with oxygen. The coefficient, under these circumstances, is only 25 per cent.

question is, what part of their oxygen carrying power do normal persons use? In table 1 values will be found for the coefficient running from 18 to 40 per cent for rest and from 44 to 64 per cent for work. Greater

variations than that could be found in the literature. During rest the Krogh-Lindhard method gives the higher coefficients. During work there is far closer agreement between the different methods, particularly with respect to the blood flow. In fact, so well do the figures for the latter agree during work that, when one considers that they are arrived at by two totally different principles, it seems reasonable to conclude that they are somewhere near the truth. Another point of interest is that with all subjects, regardless of method, the coefficient increases during work, which may be taken to mean a better economy of the circulation during work.

TABLE 1

OBSERVER	SUBJECT	METHOD	REST					WORK				
			B. F. liters per minute	Systolic output cc.	Pulse	Coefficient per cent	O ₂ absorption cc. per minute	B. F. liters per minute	Systolic output cc.	Pulse	Coefficient per cent	O ₂ absorption cc. per minute
Means and Newburgh	J. H. M.	K. L.	4.0	58	69	40	304	13.4	121	111	47	1200
Means and Newburgh	J. H. M.	Henderson	8.8	128	69	18	304	14.2	128	111	44	1200
Douglas and Haldane	Douglas	Fick	7.5	135	53	20	233	18.7	136	137	64	1974
Douglas and Haldane	Peskett	Fick	4.7	67	70	33	279	15.5	152	102	60	1653
Burwell and Robinson	Normal	Fick	3.8	63	60	25	231					

The blood flow, the coefficient, and the systolic discharge during rest remain the chief doubtful points. Y. Henderson as a result of plethysmographic studies on the hearts of animals concludes that systolic discharge is very constant. He believes that the heart delivers the same quantity of blood per beat regardless of its rate, this quantity being in humans between 1.5 and 2 cc. per kilogram of body weight. If he is right of course blood flow is directly dependent upon pulse rate. In table 1, I have shown what my own blood flow would be on his theory, taking 1.75 cc. per kilogram as the systolic output. The resting blood flow and systolic output by this method of estimation are double those by the Krogh-Lindhard method; the coefficient is half. At work, however, the values by the two methods are essentially the same.

In other words, Krogh and Lindhard and their followers believe that at rest systolic output is smaller than during work and that the resting blood flow is between 3 and 5 liters. Henderson, on the contrary, believes the resting flow to be about double that, and the volume per systole as great as during work. The Henderson theory presumes an increase in blood flow during work due solely to an increase in pulse rate, the Krogh-Lindhard an increase in flow due both to an increase in pulse rate and to an increase in the output per systole, hence a relatively greater increase in flow. The Henderson view requires a greater increase in coefficient than the Krogh-Lindhard. It is interesting to note that the results obtained by Fick's principles are divided. Those on Douglas resemble the Henderson figures, those on Peskett the Krogh-Lindhard. The rest experiment by Burwell and Robinson also agrees closely with the Krogh-Lindhard. In conclusion we may say that the resting flow is uncertain, the resting coefficient uncertain, but that the former probably lies between 3 and 9 liters per minute, the latter between 15 and 45 per cent. The flow in work is far more definite, being between 13 and 19 liters and the coefficient 40 to 65 per cent. I should add that the work in the experiments in question was of moderate severity, being equivalent approximately to climbing stairs at a rate of about 60 steps per minute.

The question now arises why is the blood flow as rapid as it appears to be? Why do we flow four or more liters at rest when it would appear that the necessary oxygen could be carried with two or less? A consideration of carbon dioxide transport and neutrality regulation will answer this question. Let us turn therefore once more to the carbon dioxide diagram. In figure 10 is shown a normal dissociation curve (Bock's) with its arterial point A. Reading off on the diagonals we find a pH for this point of 7.33.⁷ The dissociation curve for venous blood (assuming the oxygen saturation to be 65 per cent) with its venous point V^1 is shown lying just above the arterial. The pH for the point V^1 is 7.30. The amount of carbon dioxide taken up by the blood appears to be 5 volumes per cent and the increase in

⁷ pH is the logarithmic method of expressing hydrogen ion concentration. At true neutrality the pH is 7.00. As the pH rises the reaction becomes more alkaline.

carbon dioxide tension 7.5 mm. These relationships are as we actually find them in the normal subject. It will be noted that the necessary carbon dioxide is transported with only an insignificant change in blood reaction. Let us suppose that the individual whose curves we have just examined has a carbon dioxide output of 200 cc. per minute and a blood flow of four liters. Then let us suppose his

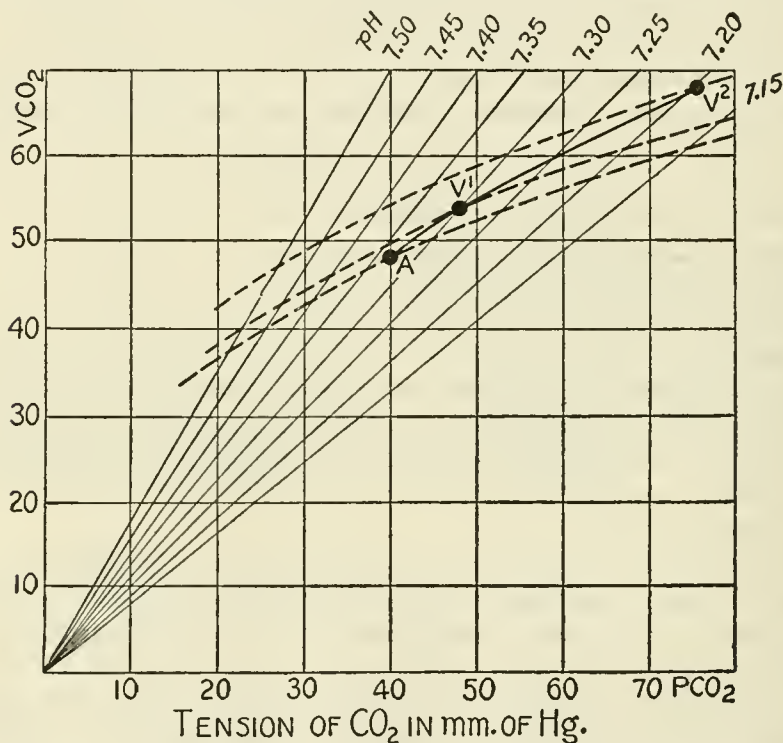


FIG. 10. THE CARBON DIOXIDE DIAGRAM SHOWING THE EFFECT OF RATE OF BLOOD FLOW UPON THE POSITION OF THE VENOUS POINT

The curves are the same as those shown in figure 4 for Bock's blood. A series of diagonals indicating the hydrogen ion concentration have been added. *A* denotes the arterial point, *V*₁ the venous point, at an assumed blood flow of 4 liters per minute, and a carbon dioxide output of 200 cc. per minute. Should the carbon dioxide output remain the same, and the blood flow drop to one liter per minute, the venous point would occupy the position shown by *V*₂. In the first instance the difference in hydrogen ion concentration between arterial and venous blood is 0.03; in the second it is 0.14. The difference in carbon dioxide tension in the first instance is 7.5 mm.; in the second it is 37 mm.

flow is reduced to one liter. It is obvious that his venous blood would have 20 volumes per cent of carbon dioxide added to it instead of 5. Under these conditions the level of the dissociation curve would rise to that of completely reduced blood as shown, but even then when we place the venous point V^2 upon it, for a level of carbon dioxide content 20 volumes per cent above the arterial, we find that carbon dioxide tension has increased to 77 mm. and that the pH has become 7.19, which is a distinct move in the direction of acidity.

It is quite apparent therefore that the slower the blood flow the greater will be the fluctuation between arterial and venous carbon dioxide tension and reaction. The slower the flow the less perfect will be neutrality regulation, the faster the more perfect. In the case supposed, with the slow flow the venous blood is definitely nearer acid than it should be, while with the higher flow there is but an insignificant change between arterial and venous. It seems fair to conclude, therefore, that although it necessitates more work on the part of the heart, a blood flow sufficiently rapid as to use but 45 per cent or less of the oxygen carrying capacity of the blood confers material benefits to the organism in the matter of neutrality regulation. Besides this it is of course altogether probable that certain organs such as the kidney require a high rate of blood flow for reasons other than those of gas exchange and the total flow may be in part kept high to meet their needs; but the stabilizing of blood reaction and the minimization of the changes between arterial and venous blood in that regard would seem to be the most fundamental determinant of the circulation's relative rate.

Pulmonary ventilation and blood flow then are both pitched at such rates as will, while taking care of the metabolic demands for gas transport, at the same time preserve blood reaction and, by providing a wide margin of safety with respect to oxygen tension, obviate any danger of asphyxia even though through muscular work or other agencies the metabolism may undergo rapid and gross alteration. Since they subserve the same purposes it would appear reasonable to suppose that their rates are similarly controlled. That carbon dioxide tension or blood reaction regulates blood flow as well as pulmonary ventilation has already been suggested by more than

one author (Y. Henderson, also Boothby). It, however, remains a suggestion thus far, although seemingly a likely one.

I shall have more to say on this general subject when we come to consider the relationships which may occur in the slow blood flow of heart disease and the imperfect oxygen carrying power of the blood in anemia and allied states.

VI. THE EFFICIENCY OF THE PULMONARY BELLOWS

We may now properly give some thought to the factors which in the beginning of the article I classified as the ones which determine the supply of ventilation. By that I meant those which affect the ability of the respiratory bellows to accomplish the task imposed upon it. This ability will depend among other things, as with any pump, upon the volume of stroke that can be delivered and the number of strokes that can be made in a unit of time.

In the case of the lungs the greatest possible stroke is but rarely employed. Nevertheless, as Peabody has shown, its magnitude bears an important relation to the functional ability of the bellows. If following the greatest possible inspiration we require a person to expel forcibly from his chest every bit of air that he can, the volume so expelled is called his vital capacity. This vital capacity does not represent the total capacity of his lungs to hold air, for after a maximum expiration, since the chest can not be completely collapsed, a certain amount of air remains in the lungs. The air so remaining is called the residual air. The vital capacity plus the residual air constitutes the total capacity of the lungs. In normal breathing at rest the whole vital capacity is not used. The subject can always expire or inspire a considerably greater volume than that of the ordinary expiration or inspiration. The amplitude of the usual tidal respiration is from 500 to 550 cc. When the chest is midway in such a normal respiratory excursion it is said to be in the midposition. The air that can be taken in by a maximal inspiration over and above the capacity at the midposition is called the complementary air and similarly that which can be expelled from the midposition is called the reserve air. Vital capacity therefore consists in complementary plus reserve air, and total capacity in complementary plus reserve plus residual air.

Numerous observers have studied the magnitude of these several factors both in health and disease. Since in the latter there may appear marked alterations it will be worth while to spend a little time examining the normal values.

The vital capacity, the complemental, reserve, and tidal air can easily be determined by causing the subject to breathe into a calibrated spirometer. To determine the residual air, however, requires an indirect method. Bohr, for example, made his subjects after a maximal expiration inspire a known quantity of hydrogen from a spirometer. Several breaths served to mix the air in the lungs and spirometer, and then by gas analysis the dilution of the hydrogen by the residual air was determined and the volume of the latter calculated. Bohr gives values for the residual, reserve and complemental air in normal persons; so also do Rubow, Peabody and Wentworth, and Lundsgaard and Van Slyke. The first three investigators got results which are in close agreement; Lundsgaard and Van Slyke got rather higher values for the residual air. This may be because their method was different. Bohr and Rubow used the hydrogen mixture method; Lundsgaard and Van Slyke used nitrogen.

In 8 normal men Bohr found the average residual air to be 1.19 liters, the reserve air 2.13 and the complemental air 2.55. Rubow in 8 normal women found the average residual air to be 1.16 liters, reserve air 1.39 and the complemental air 1.88. Lundsgaard and Van Slyke in 10 normal men found the residual air to be 1.67 liters, the reserve air 1.84 liters and the complemental air 2.48 liters; and in 7 women, residual air 1.21 liters, reserve air 1.38 and complemental air 1.80 liters.

In any given person we must have a standard of comparison for judging of the normality of any of these factors. Peabody referred his data to height and in his paper gives a set of normal standards for persons of different heights and for the two sexes. Lundsgaard and Van Slyke got a somewhat closer relation by measuring the thorax and estimating its theoretical capacity. More recently Dreyer in a very careful study of a series of normal individuals has shown fairly conclusively that vital capacity in normal persons is a simple function of their surface areas. This is not surprising for

we have noted that normally metabolism is proportional to area; so it follows that the capacity of the respiratory bellows, the duty of which is primarily to exchange the gases of metabolism, must likewise bear some relation to area. West has confirmed Dreyer's findings and has published the following standards for the vital capacities of normal persons:

Vital capacity of normal persons

	PER SQUARE METER OF BODY SURFACE
	<i>liters</i>
Men.....	2.5
Women.....	2.0
Athletes.....	2.8

These standards will serve very well for routine clinical use. The surface area can readily be obtained from the height and weight by DuBois' "height-weight chart." The vital capacity actually found in a patient divided by his surface area can be compared with the appropriate standard and expressed as a percentage variation above or below it.

As Dreyer points out, different standards will probably have to be used for different classes of individuals, for just as athletes have a relatively greater capacity than other normal persons, so too, different classes in the community will undoubtedly show different average values dependent on their occupation, physique, etc. Variations due to age undoubtedly will be found as well. Until such standards are worked out by extensive observation, the standards given by West will serve very well as a reasonably close approximation for the study of the vital capacity in disease.

Having ascertained the normality or abnormality of any given vital capacity we next wish to know whether the level of the mid-position bears a normal relation to it. This can be discovered by determining the normal relation of reserve and complemental air. We also want to know the relation of residual air to vital capacity. It is more convenient for this purpose to consider as 100 per cent the vital capacity rather than the total capacity, for oftentimes clinically it is impossible to determine the residual air, and then too

from the point of view of symptomatology it is the functional capacity that interests us rather than the anatomical.

On this basis I find that in Bohr's 8 men the reserve air constituted 45.5 per cent and the complemental air 54.5 per cent of the vital capacity and that the residual air was equal to 25 per cent more. Similarly with Rubow's women the reserve air was 42 per cent and the complemental air 58 per cent of the vital capacity, while the residual was equal to 36 per cent more. Lundsgaard and Van Slyke got similar results with their women, the reserve air being 43.5 per cent and the complemental air 56.5 per cent of the vital capacity and the residual being equal to an additional 38 per cent. With their men, however, they got different results, the reserve air being 43 per cent and the complemental air being 57 per cent of the vital capacity, while the residual was equal to an additional 39 per cent. The difference between the male series of Bohr and of Lundsgaard and Van Slyke appears to be that the former was composed of men of better physique.

After further study Lundsgaard in a recent paper gives figures somewhat different and which are, I believe, sufficiently near the truth to be accepted in the sense of standards. In our method of notation and omitting fractions which obviously are without significance, we have for normals the following:

Reserve air should be equal to about 49 per cent of the vital capacity.

Complemental air should be equal to about 51 per cent of the vital capacity.

Residual air should be equal to about 33 per cent of the vital capacity.

We may add that tidal air is somewhere in the neighborhood of 12 per cent of the vital capacity.

The next factor of prime importance in determining the supply of ventilation or the ability of the pulmonary bellows to do its task is the so-called dead space. In breathing, the total expired air is in part made up of air from the alveoli and in part of fresh air left in the dead space, that is to say the cavities of the bronchi, trachea and upper air passages. It is obvious that it is the amount of air actually reaching the alveoli, or alveolar ventilation, which counts as far as accomplishing gas exchange goes; but, on the other hand, it is the alveolar ventilation plus the ventilation of the dead space that de-

termines the magnitude of the task the bellows must do. The volume of the dead space is therefore an important factor in determining the mechanical efficiency of the bellows.

Unfortunately we have less accurate information of the size of the dead space than of that of the vital and total capacity, its measurement obviously only being possible by indirect methods. In the literature we find considerable dispute as to whether in any given individual it is a constant or a variable. The physiological and anatomical dead space are not coincident, for, as Y. Henderson has shown, gases flowing through tubes do so in an axial manner; that is to say, the forward surface of a new gas passing along a tube is not a flat surface like the piston in a cylinder but instead, because of mural friction, is in the form of a long spike progressing down the center of the tube. Thus in inspiration fresh air begins to enter the alveoli, and in expiration alveolar air issues from the nose and mouth before the anatomical dead space has been completely washed out. The dead space for oxygen is greater than that for carbon dioxide because a certain amount of the latter gas is eliminated by the mucous membrane of the trachea, bronchi and upper air passages while none of the former is absorbed by them.

The anatomical dead space was found by Loewy in a cadaver to be 144 cc. The physiological dead space in normal persons at rest has been calculated by most observers to be between 100 and 175 cc. With deep breathing Y. Henderson and also Haldane believe that there is a marked increase in the size of the dead space, not, Henderson believes, because of any active broncho-dilation, but because the bronchial tree is passively stretched with the deeper respiratory movements along with the rest of the lungs. Krogh, on the other hand, believes that depth of respiration has but little effect upon the size of the physiologic dead space.

Were there no increase in the dead space with deep breathing the most economical type of respiration, theoretically, would be that in which each single respiration was maximal, that is to say when tidal volume and vital capacity were equal. Such a type is never seen in health, and in disease only when the vital capacity is greatly reduced.

The character of the respiratory rhythm, whether it approaches a slow deep or rapid shallow type, is obviously important because

upon it may depend the total amount of pumping that must be done to satisfy a given demand for alveolar ventilation. The volume of the alveolar ventilation we have already seen is regulated chemically. That of the total ventilation will depend upon the alveolar plus whatever dead space washing the subject's type of respiration involves. This brings us naturally enough to the question of what does determine the type. In all probability the type is closely related to the innervation or nervous control of breathing as opposed to the chemical control.

The innervation of the respiratory mechanism is chiefly through the vagus nerves, which through the pulmonary plexus receive sensory fibres from the lungs themselves. The muscles of respiration fall into two groups, the intercostal which receive their motor fibres through the spinal nerves and the diaphragm which receives motor fibres through the phrenic.

The part played in the regulation of respiration by vagus borne sensory stimuli has been studied by many observers. As early as 1868 Hering and Breuer, finding evidence of inhibition of respiration when the lungs were artificially distended and stimulation when they were emptied, advanced a theory that the primary control of respiration was reciprocal stimulation and inhibition of the respiratory center through the vagi. Head in 1889 came to similar conclusions. It was not until Haldane and Priestley had demonstrated the chemical regulation of respiration that the true part played by the nervous system was understood.

F. H. Scott, who studied the relation of chemical and nervous control by the method of observing the effect of varying the composition of the inspired air both before and after section of the vagi, makes the extremely interesting suggestion that the vagi are really the great sensory nerves of the respiratory mechanism. They play the same part in the carrying out of respiratory movements that the sensory nerves from muscles and joints of an extremity do in the carrying out of a coördinated movement of that extremity. To use Scott's own words, "the vagi, in reference to the movements of respiration, must be regarded in the same light as the sensory nerves of muscle. Without the vagi the muscular movements are excessive, and thus resemble the movements of an ataxic limb." This idea

of ataxic respiration is to my mind a most apt one. After section of both vagi the alveolar carbon dioxide tension is not greatly altered, nor is the alveolar ventilation, but the respiration rate becomes very slow and the volume of each respiration very much greater. The respirations are slow in rate but excessive in amplitude. This type of breathing seems to be the intrinsic or basic type of the respiratory center. It is the type that occurs when the center receives chemical but no nervous stimuli. It reminds one very strikingly of the slow intrinsic rhythm of the heart ventricles when they are cut off from the pacemaker in complete heart block; in fact, one might almost call the respiration after section of the vagi, respiration block.⁸

In the normal organism the increased ventilation with increased carbon dioxide content in the inspired air is accomplished by an increase in both rate and amplitude. After section of the vagi, though the reaction follows a more or less normal curve, the increase is met by an increase in amplitude only, the rate remaining constant. As Bayliss says,

It would appear, then that the inspirations excited by carbon dioxide in the normal state are cut short by the vagi inhibiting the discharge of the center; collapse of the lungs follows rapidly, the inhibition ceases, and the center is again accessible to excitation by carbon dioxide. In this way we have the advantage of an increased rate in addition to an increased depth. It is interesting that the excitatory effect of carbon dioxide does not cause the center to discharge more frequently, but with increased strength of discharge. The function of the nervous regulation is thus to moderate the discharge, which tends to be "all or none," in the absence of inhibitory impulses.⁹

We may sum up then by saying that the nervous control of breathing plays no part in the regulation of the volume of the alveolar ventilation, but that it does play a very definite part in determining the type of breathing that is to accomplish the amount of alveolar ventilation chemically required. It follows from this that nervous control plays a part in determining the total volume of the ventilation

⁸ Since writing this I have found a similar comparison by Ff. Roberts.

⁹ Recent work by Lumsden shows that the so-called respiratory center is actually four centers with somewhat different functions.

or the size of task that the bellows must do. It is apparent therefore that in disease when vital capacity may be diminished, dead space increased, or the demand for ventilation increased, the type of breathing as determined through nervous channels becomes of clinical importance.

The possible extremes of respiratory type are limited on the one hand by the vital capacity and on the other by the size of the dead space. A type in which each breath is maximal, that is equal to the vital capacity, would require a minimum respiratory rate. As the volume of the tidal excursion diminishes respiration rate must increase, but it is obvious that if the tidal excursion ever shrank to be no greater in volume than the dead space even an infinitely fast rate of breathing would yield no ventilation of the alveoli.

The respiratory type of normal persons seems to be a happy medium determined, as I have said, in all probability through nervous channels. Though a type of respiration in which each breath were maximal might theoretically seem to be an economical style of breathing, in point of fact it is quite the reverse because it necessitates a greater muscular effort than the usual type. This is because although inspiration is always an active muscular process, expiration is not. After active inspiration has been inhibited by vagus action the chest collapses passively, expelling the tidal air (450 to 550 cc.). To expel the reserve air requires an active expiration. With the usual sized tidal excursion, therefore, only one muscular act is necessary per breath; with abnormally large tidal excursions, two. Furthermore one can easily prove to himself that it requires more effort to push out the last portions of the reserve air than it does the first. Therefore, within limits we may say that as the volume of the tidal excursion approaches the vital capacity, the greater will become the muscular effort required per breath. In the reverse direction, however, the more nearly the tidal volume approaches the dead space, the greater will be the total pumping required for a given task. Respiratory types of either extreme therefore are undesirable and the usual medium type is truly a happy medium. In general the nervous control of breathing may be expected to secure for us that respiratory type which, in accordance with the local condition in our chest or abdomen, may most comfortably supply the ventilation required

for the gas exchange which our general bodily processes may at any time happen to demand.

In closing this phase of the subject I want to give briefly some idea of the actual volume of the alveolar and total ventilation in normal human beings under basal conditions. Since expired air is merely alveolar air diluted with dead space air it can easily be seen that the difference in carbon dioxide content between them will express the ratio of alveolar to dead space ventilation. It is also apparent from what has gone before that the relative amount of dead space ventilation is dependent upon the type of breathing and the size of the dead space.

The volume of the alveolar ventilation we have already noted is dependent on alveolar carbon dioxide tension and carbon dioxide output, that is to say, upon metabolism, blood flow and blood bicarbonate level. The carbon dioxide output of a normal man 165 cm. high and weighing 60 kgm., under basal conditions, would be about 185 cc. per minute. If his carbon dioxide tension were 44 mm., his alveolar ventilation would be 3 liters per minute. A man 185 cm. in height and weighing 80 kgm. similarly would have a carbon dioxide output of 227 cc. per minute, and if his alveolar carbon dioxide tension were 37 mm. his alveolar ventilation would need to be 4.4 liters per minute. In general, then, the alveolar ventilation of normal adults when under basal conditions will vary between 2.5 and 5 liters per minute.

The total ventilation is as much greater than this as its carbon dioxide content is lower than that of alveolar air. We may take from 5 to 6 per cent as the normal range of alveolar carbon dioxide content. That of the expired air we can gain some notion of by reference to any of the extensive respiration experiments recorded in the literature. For example, in a series of seventeen normal men Carpenter and collaborators found an average carbon dioxide content in the expired air of 3.67 per cent and the extreme variations above or below this average were 4.29 and 3.06 per cent. The man with the highest carbon dioxide content had the slowest respiration rate in the group and consequently diluted his alveolar air with dead space air the least number of times per minute, and conversely the man with the lowest carbon dioxide content had the most rapid respira-

tion. Benedict and coworkers, in a series of thirteen other normal men, got an average carbon dioxide content of 3.34 per cent in the expired air, the maximum in the series being 3.67 per cent, the minimum 2.83 per cent. The total ventilation therefore of normal adults under basal conditions may be expected to vary between 4 and 8 liters per minute. The carbon dioxide content of the expired air would be the proper criterion for judging whether any given individual's ventilation is abnormally high or low, provided it is at the same time compared with that of his alveolar air.

VII. DYSPNOEA ASSOCIATED PRIMARILY WITH INCREASED METABOLISM

We are now in a position to discuss the symptom dyspnoea more directly. As I pointed out at the beginning of the paper dyspnoea is primarily a symptom, a subjective phenomenon. Increased pulmonary ventilation which is not distressful is not dyspnoea; it is better termed hyperpnoea. Usually hyperpnoea precedes dyspnoea, but not necessarily, for occasionally in disease we meet with breathlessness when there is no actual increase in pulmonary ventilation. In this section we will consider the hyperpnoeas due mainly to increased rate of metabolism and the extent to which they may produce dyspnoea.

The most potent accelerator of metabolism, as said before, is the performance of muscular work, and it gives rise to the greatest degree of hyperpnoea that we ever encounter. In the normal man the hyperpnoea induced by muscular work finally reaches a level at which the respiratory organs meet with some embarrassment in providing the necessary ventilation; then dyspnoea results. Dyspnoea therefore may be physiologic as well as pathologic.

Let us consider physiologic dyspnoea for a few moments. The effect of muscular work on the external respiration and the gas exchange has been studied by a large number of investigators. Even as far back as 1789 Sequin and Lavoisier made observations of the oxygen intake and carbon dioxide output of human subjects during work. Pettenkofer and Voit in Germany, Haldane and his school in England, and Benedict and his coworkers in this country have all made extensive observations of the gas exchange during work.

From these studies it may be said that during muscular work there is a steady rise in oxygen intake and carbon dioxide output, that is in metabolism, as the amount of work increases. The pulmonary ventilation also increases in direct proportion to the carbon dioxide output in accordance with Haggard and Henderson's first law of breathing. During heavy muscular work on a stationary bicycle, the subject being a professional bicycle rider, Benedict and Cathcart found oxygen absorption as high as 3 liters per minute, their subject's resting or basal oxygen absorption being only about 250 cc. per minute. The increase in the gas exchange during work therefore rose to over ten times the resting value.

The percentage increase in gas exchange over the resting value for any definite amount of external work can not be foretold. It will vary with different individuals and will be dependent upon the relative efficiency of the individual as a machine.

Benedict and Cathcart, for example, in several subjects who were not trained athletes, found that the net efficiency during muscular work was between 20 and 23 per cent; that of the trained bicycle rider was 25 per cent. By net efficiency is meant the ratio of the heat equivalent of the actual work performed to the increase in heat production over the subject's basal metabolism during the performance of the work. It is precisely the same ratio that is used in estimating the efficiency of such a machine as a locomotive when the fuel value of the coal burned is compared with the heat equivalent of the work actually accomplished. Benedict and Cathcart's athlete, being a more efficient machine than the non-professional subjects, was able to do the same amount of work as they with a somewhat lower actual metabolism. Training, then, undoubtedly enables one through improved coördination and muscular adaptation to do muscular work with increasing efficiency, to accomplish a given result with a decreasing expenditure of energy.

The amount of external work necessary to produce dyspnoea therefore may be expected to vary among individuals in accordance with their net mechanical efficiency. But aside from this, even with like metabolism elevation and like degree of hyperpnoea, the point of onset of dyspnoea will vary among different individuals. Thus Peabody has shown that the appearance of the symptom dyspnoea

is more closely related to vital capacity than to the degree of hyperpnoea. Persons with low vital capacity will become subjectively breathless, that is dyspnoeic, with comparatively slight degrees of hyperpnoea, while those with large vital capacities will be able to increase their ventilation to a marked degree before being subjectively aware of the increase. Grabfield and I, in some rebreathing experiments on ourselves, found that we were aware of no increase in breathing until the ventilation had been approximately doubled and had no real discomfort, dyspnoea, until it had been increased four- or fivefold.

The point in the ventilation curve at which breathlessness first appears and the point at which dyspnoea becomes intolerable to the subject, in other words the point at which he has to quit work and "recover his breath," will depend, if not entirely, certainly at least in large part, upon the mechanical efficiency of his pulmonary bellows. This efficiency can be greatly increased by athletic training. Just as vital capacity can be increased by respiratory exercises, so can the mechanical ability to increase lung ventilation be improved through training. An athlete may run a quarter mile race with but little dyspnoea, while an untrained man of the same size running at the same speed might be quite "blown" at the end of an eighth of a mile. The relative hyperpnoea of the two subjects would probably not be very different but the athlete with efficient bellows would have little discomfort while the untrained subject would have considerable. Of course it might be argued that the difference between trained and untrained subjects is circulatory rather than ventilatory. I dare say it is both, but I believe the weight of evidence points to the latter as the most important as between individuals with normal hearts.

Another factor which may play some part in determining differences between individuals is the rate of diffusion of gases through the lungs. Harrop, using Maria Krogh's method, has recently shown that there is considerable individual difference in this rate among normal persons. Those with the less permeable lungs will have dyspnoea sooner than those with the more permeable.

It is fortunate perhaps that there is even in health such a symptom as dyspnoea, for were there not it is conceivable that muscular work might be carried to the point at which either pulmonary ventilation

or blood flow would become grossly insufficient so that the subject might become seriously acidotic or drop dead from asphyxia. The development of dyspnoea prevents this, however, and may therefore be regarded as a protective phenomenon. In the normal subject, at least, long before any such danger level is reached dyspnoea will become intolerable and muscular work accordingly will be either decreased or abandoned.

In this connection and also with reference to the remarks in the preceding section on the relation between the chemical and nervous control of breathing, the studies of Krogh and Lindhard on the various factors of respiration during the sudden transition from rest to hard work are of interest. These investigators found, in brief, that when work is suddenly started there is an almost instantaneous rise in pulmonary ventilation, pulse rate and blood flow. This rise was too sudden to be due to the chemical stimulation of the respiratory center for it occurred before any increased carbon dioxide tension or pH could have reached the medulla via the blood stream. It must therefore have been due to a nervous mechanism. They concluded that it was not a reflex but rather that it was due to the irradiation of impulses from the motor cortex and the stimulation of the medullary centers thereby, for they found that when subjects were told to begin work and expected a heavy load the same increase in ventilation and blood flow occurred even though there actually was no load on the work apparatus. It was, in other words, the mental effort to work that caused the reaction, not the actual work done.

The same writers more recently have studied respiration during the transition from work to rest. The effect here was a gradual one. The hyperpnoea did not cease abruptly when work was discontinued but instead, rather gradually. This finding is consistent with the common knowledge that dyspnoea induced by work may persist for varying lengths of time after the work has ceased. This matter will be discussed further in a later section.

We now come to dyspnoea associated with hypermetabolism induced by disease. The two afebrile disease states outstandingly characterized by an elevated metabolism are hyperthyroidism and leukemia.

Let us consider the ventilatory problem that confronts the patient with exophthalmic goiter. This can be perhaps visualized more readily if we make use of a simple diagram. In figure 11 the construction is based on the work of various writers. It is merely an

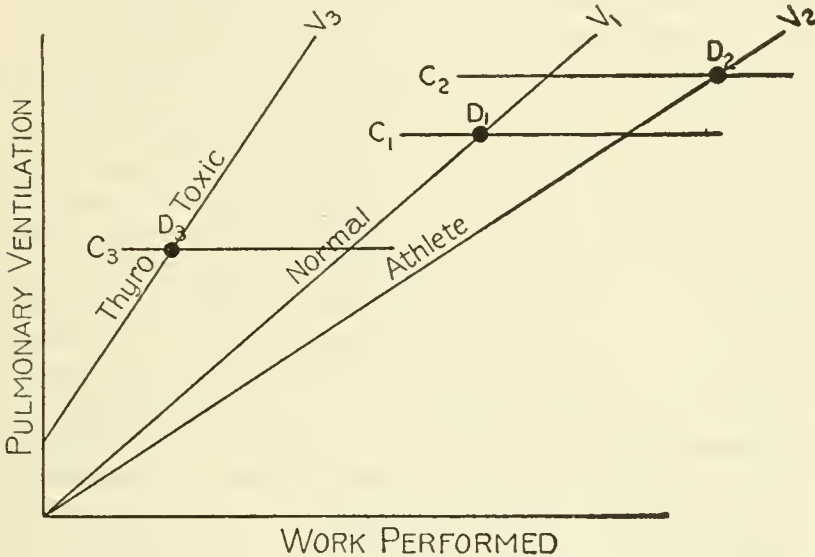


FIG. 11. SCHEMA SHOWING DYSPNOEA POINTS IN DIFFERENT TYPES OF PERSONS

The line V_1 denotes the ventilation reaction to work in a normal person (Means and Newburgh); the line C_1 the level of hyperpnoea which produces dyspnoea (Grabfield and Means). D_1 is therefore the dyspnoea point. V_2 denotes the ventilation reaction of an athlete. It is less steep than V_1 because, as Benedict and Cathcart have shown, athletes are more efficient machines. The line C_2 is drawn higher for the athlete because West has shown their vital capacity is greater than normal.

V_3 denotes the reaction in a thyrotoxic person. It starts at a higher level than V_1 and V_2 because of the elevated basal metabolism. It is steeper than V_1 because, as Plummer and Boothby have shown, thyrotoxic persons are less efficient machines than normals. The line C_3 is drawn at a lower level than C_1 because Rabinowitch has demonstrated a marked reduction in vital capacity in thyrotoxicosis.

The abscissae of points D_1 , D_2 , D_3 indicate the amount of work that can be done by the three types of persons up to the point of dyspnoea.

approximation and is by no means intended as a quantitative expression of what takes place. Accepted as schematic only it may serve to clear up certain points which might be confused. Let the diagonal line V_1 represent the ventilation response of a normal individual to

increasing amounts of muscular work. The ordinates represent percentile increase in pulmonary ventilation, the abscissae the amount of muscular work done. The line C_1 is drawn at the level at which the hyperpnoea may be expected to produce dyspnoea, that is to say, when the ventilation has been increased in the neighborhood of four- or fivefold the resting value. D_1 , the point at which C_1 intersects V_1 , may be taken as the dyspnoea point for the normal individual and its abscissa the rate of work that will cause actual dyspnoea. The level of C_1 will be closely related to the vital capacity. In athletes the dyspnoea point will be raised for two reasons: in the first place, their vital capacity being relatively greater, we may expect that the degree of hyperpnoea required to produce dyspnoea will be raised, C_2 for example; also, from Benedict and Cathcart's work, we may expect them to be more efficient machines. This means that for a given rate of work less energy exchange and hence less ventilation will be required of them. The ventilation response of the athlete then will approach the position of V_2 rather than V_1 and his dyspnoea point will fall in some such position as D_2 . In other words, he can perform work at a higher rate than the non-athletic normal subject without becoming dyspnoeic.

The situation in toxic goiter I fancy to be something as follows: in the first place, in this disease even at rest the metabolic rate is elevated. The line representing ventilation response to work therefore, instead of starting at the base line, will start somewhat above it, as is shown by V_3 . Were the thyrotoxic individual equally efficient with the normal, the line V_3 might be expected to be parallel to V_1 . Plummer and Boothby, however, have shown that the net efficiency of such patients is less than that of the normal; therefore we may expect the ventilation response to approximate more closely the slope of V_3 than of V_1 . With regard to the amount of hyperpnoea necessary to produce dyspnoea, the work of Rabinowitch is of importance. This author finds a definite decrease in vital capacity in toxic goiter, the degree of decrease being proportional to the degree of hyperthyroidism. In the diagram I have therefore drawn the line C_3 representing a lower level of hyperpnoea necessary to produce dyspnoea based on Rabinowitch's finding. The dyspnoea point of the thyrotoxic person may be expected to lie in some such position as D_3 .

This diagram makes it apparent that the dyspnoea of toxic goiter can in large measure be explained on the basis of increased metabolic rate together with that of decreased vital capacity and decreased bodily efficiency. The thyrotoxic individual, in other words, becomes dyspnoeic more readily than the normal (1) because of the handicap of his increased basal gas exchange, (2) because it takes a greater expenditure of energy to accomplish a given task and hence a relatively greater degree of hyperpnoea, and (3) because he has a less efficient pulmonary bellows. It is not necessary to assume any circulatory insufficiency to account for the dyspnoea. It can be accounted for amply without this. I do not mean to imply by this that there never is any cardiac element in toxic goiter. It is a matter of common knowledge that frequently there is, but my conception is that when present it is to be regarded as an added factor causing dyspnoea, not the primary factor. The primary factor is the increased metabolic rate.

The remarks made so far about work, hyperpnoea and dyspnoea in the normal and thyrotoxic individual apply to continuous work being done at a constant rate. The matter of sudden exertion is somewhat different. I have mentioned Krogh and Lindhard's findings on this latter point. They found, it will be recalled, an instantaneous hyperpnoea even on the mere expectation of exertion. In thyrotoxic persons this phenomenon may be exaggerated even to the point of producing dyspnoea.

The duration of dyspnoea after cessation of work is also of interest. In a research now in progress at the Massachusetts General Hospital, S. G. Mudd has found that after doing a standard piece of exercise not only is there a greater percentile increase in ventilation over the resting value in thyrotoxic than in normal individuals but also that the former require a considerably longer time to recover from the effect.

The dyspnoea of thyrotoxicosis is obviously a dyspnoea on exertion only. It occurs when the extra calories of work are added to those due to the thyrotoxicosis. To account for dyspnoea at rest solely on the basis of increased metabolic rate we should have to assume an increase of two or three hundred per cent, but such has never been

observed. When in a thyrotoxic state real dyspnoea occurs at rest I believe we should expect a different cause, usually to be found in a damaged heart.

VIII. DYSPNOEA ASSOCIATED PRIMARILY WITH DISTURBANCE OF ACID-BASE BALANCE

Just as from Haggard and Henderson's first law of breathing it followed that increased metabolism will produce hyperpnoea, and therefore the possibility of dyspnoea, so does it likewise from their second law that acidosis will do the same.

The term acidosis is much used and much abused, not only by clinicians but by physiologists. As a start it is necessary that we seek a clear definition. It is known that considerable quantities of non-volatile acid may be added to the blood stream of warm blooded animals without greatly altering the normal ratio of acid to base, that is to say, the hydrogen ion concentration. It is also generally understood that the compensating mechanism in preserving neutrality is first increased ventilation of the lungs, hyperpnoea, and consequent washing out of carbon dioxide, and later increased production of alkali in the form of ammonia and increased elimination of acid by the kidneys. The reaction of the blood does not change materially on the addition of non-volatile acid because by the liberation of carbon dioxide alkali is set free to combine with the new acid.

We may therefore perhaps define acidosis as a condition in which acids other than carbonic are present in the blood in excess of their usual concentration. We may expect such a state to be accompanied by (1) a lowered blood bicarbonate level, (2) hyperpnoea, (3) lowered alveolar carbon dioxide tension and (4) lowered blood carbonic acid concentration. It is not accompanied by any material increase in hydrogen ion concentration of the blood unless the compensating mechanism (hyperpnoea) fails.

The sequence of events which may be expected to happen upon the addition of acid are as follows: in the first place we have the normal situation

$$[\text{H}^+] = K \frac{[\text{H}_2\text{CO}_3]}{[\text{BHCO}_3]}$$

Under these circumstances, $\frac{[\text{H}_2\text{CO}_3]}{[\text{BHCO}_3]}$ has a value of about $\frac{1}{20}$ (while $K = 7 \times 10^{-7}$) therefore we may write

$$[\text{H}^+] = K \frac{1}{20}$$

The free carbonic acid is represented by the numerator of the fraction, 1, and the bicarbonate by the denominator, 20. Suppose now sufficient non-volatile acid were added to the blood to combine with one-half the bicarbonate. The formula would then become

$$[\text{H}^+] = K \frac{1 + 10}{10}$$

for ten units of carbonic acid would be set free from ten units of bicarbonate decomposed and therefore would move from the denominator to the numerator of the ratio. No such degree of change as this actually occurs in vivo, for as fast as the denominator is decreased the $[\text{H}^+]$ is increased; this stimulates the respiratory center, hyperpnoea ensues, carbon dioxide is washed out and the numerator is reduced sufficiently to preserve the normal ratio. In this instance we should have

$$[\text{H}^+] = K \frac{1 + 10 - 10.5}{10}$$

or

$$[\text{H}^+] = K \frac{0.5}{10}$$

which is a completely compensated state in which blood reaction is normal and alveolar carbon dioxide tension reduced one-half and by the same token pulmonary ventilation doubled.

This set of reactions is of interest because it shows not only the importance of the rôle which carbon dioxide plays in the maintenance of blood neutrality, but also why the respiratory center and pulmonary ventilation are more obviously adjusted in accordance with the requirements of carbon dioxide elimination rather than those of oxygen intake. It would also seem to lend weight to the view that hydrogen ion concentration, not carbon dioxide tension per se, pro-

vides the true stimulus to the respiratory center, for were it the latter the reaction might be expected to stop at

$$[H^+] = K \frac{1}{10}$$

instead of going on to

$$[H^+] = K \frac{0.5}{10}$$

After the addition of non-volatile acid and compensation through the lungs to the point expressed by $[H^+] = K \frac{0.5}{10}$, restoration to the original state of $[H^+] = K \frac{1}{10}$ can only be brought about by the elimination of non-volatile acid by the kidneys and the reaccumulation of fixed base.

It is not within the scope of the present paper to elaborate upon neutrality regulation. This has been covered in the monographs of Van Slyke and of Wilson. We are concerned now with the relation of acidosis to dyspnoea. In considering this relationship the important thing for the clinician to remember is that it is acid that stimulates the respiratory center; normally H_2CO_3 is the chief acid involved but other acids such as betaoxybutyric or lactic will act in a similar way, and therefore their presence in the blood will produce hyperpnoea.

The hyperpnoea of the acidotic individual, like that of the thyrotoxic individual, will become dyspnoea when the pulmonary bellows meets with embarrassment in the delivery of the ventilation that the disturbance of acid-base balance demands.

We meet with acidosis clinically under a variety of circumstances. For example, there are metabolic acidoses, the over-production of non-volatile acid as in diabetes, retention acidoses as in nephritis when the kidneys may fail to eliminate the non-volatile acid products of normal metabolism with the result that they pile up in the blood, and finally alimentary acidoses, due to the ingestion of acid of which methyl alcohol poisoning is a fair sample.

The diabetic with acidosis but rarely has what would truly be called dyspnoea, at least at rest. The characteristic type of breathing in that condition is slow and deep but easy; only in the very

severe grades does it become labored. It will be recalled that in the normal hyperpnoea did not give rise to real dyspnoea until the ventilation had been increased four- or fivefold. Such a degree of hyperpnoea would result only when the bicarbonate of the blood had been reduced to about twelve volumes per cent and the carbon dioxide tension to about 10 mm. A glance at the carbon dioxide diagram will show that this would be a very low level curve. In the diabetic there is no reason to expect a reduction of the dyspnoea point for there is no obvious reason for impairment of the pulmonary bellows. Therefore, in diabetic acidosis we may have what may be called a pure hyperpnoea, as in moderate muscular work in the normal person, with true dyspnoea only in the most severe grades, that is to say when more than two-thirds of the blood bicarbonate has been put out of commission through union with acid. It must be pointed out too that when the acidosis reaches that grade of severity the patient is usually in coma, so that the breathing would be labored objectively only. Of course in milder grades of diabetic acidosis there may be dyspnoea on exertion as there is in the thyrotoxic person.

The nephritic with acidosis is much more likely to have dyspnoea, this being because along with the acidosis there is usually a definite handicap in the way of ventilation due to reduced vital capacity from pulmonary edema or congestion, and very likely an insufficient circulation which later we shall see is a cause for dyspnoea per se. Retention acidosis occurs in chronic nephritis only when there is very severe impairment of renal function, generally as a terminal phenomenon and coincident with cardiac failure, uremia or both.

In studying the various types of acid-base balance that may occur clinically, the carbon dioxide diagram is most helpful. Using Haggard and Henderson's method of plotting Van Slyke has pointed out that there are eight types of abnormality at least theoretically possible. In figure 12 I have shown two shaded zones, one denoting the levels within which the carbon dioxide dissociation curves of normal bloods fall (*C*), the other (*pH*) the range of normal blood hydrogen ion concentration. The arterial or *A* points of all normal bloods should fall within the double shaded lozenge formed by the intersection of these zones, position *I*. If acid is added to the blood stream the curve level will fall, but so long as pulmonary compensation

is adequate, that is to say so long as sufficient hyperpnoea develops to affect the new acid through reduction in carbonic acid, the A point will still fall within the zone pH, in position 2. If, however, hyperpnoea is not sufficient we shall have what may be called an

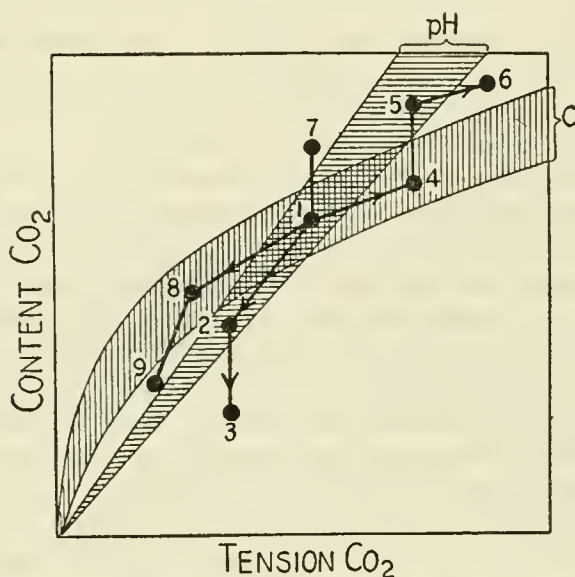


FIG. 12. DIAGRAM SHOWING THE SEVERAL POSSIBLE VARIETIES OF ACID-BASE BALANCE DISTURBANCE

The ordinate denotes carbon dioxide content; the abscissa, carbon dioxide tension. The perpendicularly shaded zone C is that in which carbon dioxide dissociation curves of normal persons fall. The horizontally shaded triangle shows the range of the pH of normal blood. The doubly shaded diamond (1) denotes the locus of normal arterial points. Points 2 to 9 show loci under abnormal circumstances, these being all possible combinations of increased, decreased or normal pH with increased, decreased or normal curve levels. There is probably not enough data yet in the literature to give accurate limits to the two zones shown. This diagram is to illustrate the principles involved and not the exact magnitudes of the several factors concerned. The locus of normal arterial points of course does not really form a diamond. It is much more likely to be an oval shaped area.

uncompensated acidosis. Blood reaction will actually shift in the direction of acidity and the A point will move to position 3. It should be remembered in studying such diagrams that shifts to the right are in the direction of acidity, to the left in that of increased alkalinity.

Other disturbances may occur clinically. Suppose that although no abnormal acid is present in the blood, ventilation becomes insufficient. Under these circumstances there would be no downward movement of the curve, but there would be an increase in carbon dioxide tension due to an insufficient rate of elimination of that gas. The *A* point would move to position 4, which although not an acidosis in the way we have defined it, still would be a state in which the blood reaction is shifted in an acid direction. It might very suitably be called a carbonic acidosis. The compensation for such a state would lie in the accumulation of a supernormal blood bicarbonate concentration. The curve level would then rise until without necessitating any change in carbon dioxide tension (and hence in pulmonary ventilation) the *A* point would come to fall in the normal pH zone, position 5. If compensation again failed it would move to position 6 and we should once more have carbonic acidosis, this time with a high level curve.

The conditions in which the blood reaction is shifted in the alkaline direction do not directly concern us here, yet they may profitably be mentioned in passing. If large doses of alkali be given and if ventilation be not correspondingly reduced we should have the condition of alkalosis, position 7. If, on the other hand, we voluntarily increase our ventilation, as has already been seen, carbon dioxide will be washed out, carbon dioxide tension will be reduced and the *A* point will move to position 8. If this forced breathing is continued further, Henderson and Haggard have shown, a compensatory lowering of the dissociation curve occurs. This they believe is due to the withdrawal of base by the kidneys and more particularly by the tissues. When such compensation has taken place the *A* point moves to position 9, and theoretically it might even go on to position 2. These changes have been produced by Grant and Goldman in themselves. The condition from the point of view of blood reaction is one of alkalosis, yet with a low level curve. It is what Henderson calls acapnia.

The diagram shows clearly that neither carbon dioxide tension alone nor blood bicarbonate alone can tell us the true state of acid-base balance. In all of the states denoted by positions 2, 3 and 9 there is a lowered carbon dioxide tension and also a lowered

bicarbonate, yet 3 is in a sense the antithesis of 9. The whole problem is one of which is cause and which effect. Is the curve low as a direct result of the primary abnormality or to compensate for it? Fortunately it is usually possible to settle this point by inference. If hyperpnoea develops in the diabetic or nephritic, or if their blood bicarbonate or carbon dioxide tension is found lowered it is safe to assume that there is true acidosis, not acapnia. Under such circumstances either alveolar or blood carbon dioxide alone will give valuable information as to the severity of the acidosis and certainly is sufficient for clinical routine. In other conditions such as shock or ether intoxication with lowered curve levels, the problem of what is cause and what effect is far more difficult, in fact even the complete carbon dioxide diagram may not tell, for as noted, position 2 theoretically at least might be arrived at from opposite directions. To the various possible disturbances of acid-base balance Van Slyke has given descriptive terms. For most purposes the simple designation of normal, increased or decreased hydrogen ion concentration with normal, high or low level curve as the case may be, will suffice.

In concluding this phase of the subject it may be of interest to examine the actual findings in certain acidotic individuals. We will take first the case of L. H., a boy with severe diabetes. On January 23 he had a severe acidosis as shown by a low position of the carbon dioxide dissociation curve, but with good compensation as shown by the position of the *A* point (fig. 13). He was not in coma but was distinctly drowsy. His alveolar carbon dioxide tension was 14.6 mm. and at that tension the carbon dioxide content of his arterial blood was 16 per cent. Spirographic tracings showed that his total ventilation under these circumstances was 10.6 liters per minute, his respiration rate 11.2 per minute and the volume of his tidal air 950 cc. On January 27 he was much better. His alveolar carbon dioxide tension was 26.8 mm. and at that tension the carbon dioxide content of his arterial blood was 36 volumes per cent. His total ventilation on this day was only 4.5 liters per minute, his respiration rate was 10 per minute and the volume of the tidal air 450 cc. Approximately, when his carbon dioxide tension was doubled, his ventilation was halved, thus bearing out Haggard and Hen-

derson's second law. It is also of interest to note that the hyperpnoea of the first observation was secured largely by an increase in amplitude rather than in the rate of breathing. The hyperpnoea which, as I have said, was about twice his normal ventilation gave rise to no subjective dyspnoea, which is what we should expect. Three carbon dioxide diagrams were obtained in this case and are shown in figure 13. They illustrate the sequence of events in a

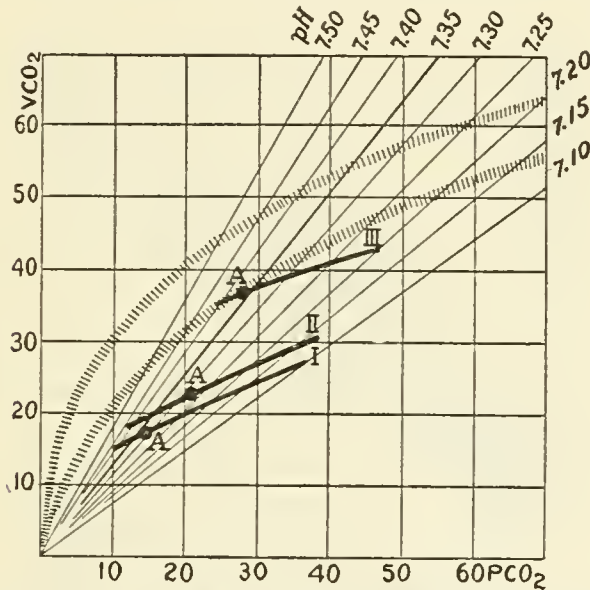


FIG. 13. THE CARBON DIOXIDE DIAGRAM IN DIABETIC ACIDOSIS

Patient L. H. Curve I was obtained upon January 23, curve II on January 24 and III on January 27. The shaded zone represents that in which the curves of normal persons are believed to fall.

metabolic acidosis of moderate severity and the gradual return toward a normal state during treatment (dietetic). A much more complete set of observations of this sort by Bock, Field and Adair has recently appeared.

In this connection a tracing published by Rogers and myself, which was of one of the most severe acidosis hyperpnoeas I have ever seen, is of interest. The patient, a negro of forty-six years, had congenital cystic kidneys. The tracing was obtained two

days before death. His renal function was very low and the excretion of acid greatly impaired. He was suffering, in other words, from a retention acidosis. Whether it was compensated or not we cannot say for no carbon dioxide diagram was obtained; on the chances it was not. At any rate the carbon dioxide capacity of the plasma was greatly reduced and his alveolar carbon dioxide tension was only 6.4 mm. The total ventilation per minute was 55 liters,

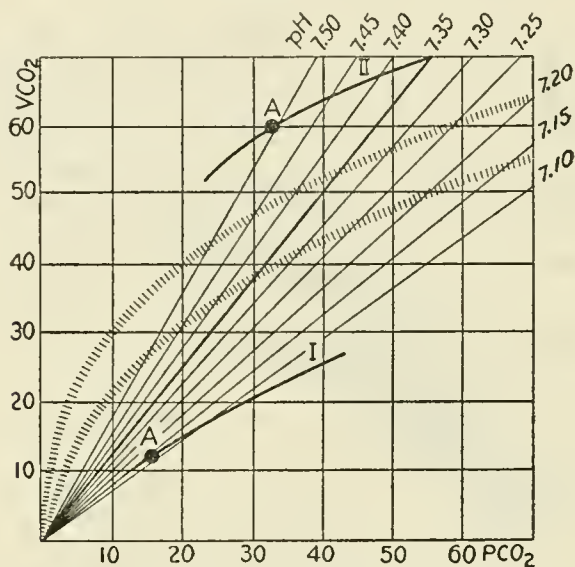


FIG. 14. THE CARBON DIOXIDE DIAGRAM IN RENAL ACIDOSIS

Curve I was obtained on February 19. Patient had a very severe acidosis. At the time he had intense hyperpnoea. Curve II was obtained the next day after the administration of large quantities of sodium bicarbonate and shows a condition of alkalosis, the hyperpnoea was gone.

the respiration rate 22.5 per minute and the volume per respiration 2440 cc. There was very definite dyspnoea.

Another interesting case of retention acidosis is that of a man of fifty-five who was brought to the hospital with an intense grade of hyperpnoea and marked dyspnoea. His underlying malady was arteriosclerotic nephritis but he had in addition an obstructing prostate and double pyelonephritis. He was in a profound state of uremia and acidosis. The carbon dioxide diagram obtained on this

day is shown in figure 14, curve I. The curve is very low, indicating that the blood bicarbonate was greatly reduced. The position of the *A* point moreover shows that there was a shift in blood reaction in the acid direction. The acidosis was not fully compensated for; great as his hyperpnoea was, it was yet not great enough to preserve the normal ratio between acid and base. Curve II (fig. 14) shows the state of affairs on the following day. After large doses of alkali (too large as the diagram shows) not only was the acidosis completely abolished but he was actually rendered alkalotic, that is to say there was decreased hydrogen ion concentration with a high level curve. The amount of sodium bicarbonate given between these two observations was approximately 175 grams. If we turn back to figure 12 it will be seen that his arterial point travelled in twenty-four hours from position 3 to position 7. No less remarkable was the change in his clinical appearance. The hyperpnoea, and with it the dyspnoea, had entirely gone and his mental state had changed from a state just bordering on coma to one of normal alertness. Needless to say the improvement was of short duration.

It is of interest that before the days of the carbon dioxide diagram Barcroft and his coworkers had found evidence of retention acidosis in cardio-renal disease. Their method of approach was through the behavior of the oxygen dissociation curve instead of the carbon dioxide. The interdependence of the two curves I have already emphasized, and incidentally it should be pointed out here that the phenomena of the oxygen curve had been extensively studied by Bohr and by Barcroft several years before Haldane and his coworkers discovered those of the carbon dioxide curve. Concerning disturbances of acid-base balance much the same information can be gathered from the oxygen curve as from the carbon dioxide, but since studies of the latter are somewhat easier to carry out, for most purposes they are likely to supersede those of the oxygen curve.

With acid added to blood the oxygen curve moves to the right just as the carbon dioxide curve; and with alkali to the left. With the oxygen curve, however, carbon dioxide has the same effect as any other acid; it shifts the curve quite as much as an equal concentration of non-volatile acid. This amounts in brief to the following: the carbon dioxide tension remaining constant, at any given tension of oxygen,

blood to which a non-volatile acid has been added will take up less oxygen than normal blood; also, at any given tension of oxygen, normal blood will take up progressively less oxygen as carbon dioxide tension rises.

The condition of the blood in which it takes up less oxygen than normal blood at a given tension of oxygen Barcroft calls *meionexy* and such blood *meionectic*. The reverse process happens upon the addition of alkali; the curve moves to the left and at a given tension of oxygen takes up more oxygen than normal blood. This condition he calls *pleonexy* and such blood *pleonectic*. Blood with the curve in the normal position he calls *mesectic*.

In 1913 Barcroft and others, in seeking a cause for the dyspnoea occurring in cases of cardio-renal disease unaccompanied by cyanosis, found in some of them not only a potential *meionexy*, as shown by a diminished oxygen capacity of the blood when exposed to a fixed tension of oxygen and in the absence of carbon dioxide, but also in some an actual *meionexy*, as shown by a *meionectic* position of the dissociation curve when the blood was exposed to the patient's actual alveolar carbon dioxide tension. Barcroft thought that this *meionexy* explained the dyspnoea and that "there is no further mystery about it." In a sense he is probably right. Acidosis produces hyperpnoea and he had furnished proof of acidosis. As we have seen, however, the point at which hyperpnoea gives rise to dyspnoea is dependent on a variety of pulmonary and circulatory factors, so the relationship is not perhaps as simple as Barcroft fancied.

Inspired by Barcroft's work and to get further information on this interesting subject I undertook in 1914 a similar study. I was able to confirm his finding that acidosis may occur in cardio-renal disease. The method employed was the determination of the oxygen saturation of the blood at a fixed tension of oxygen (17 mm.) and in the absence of carbon dioxide. Under such circumstances normal blood becomes from 60 to 75 per cent saturated; acidotic blood less saturated, the greater the acidosis the less the saturation. This procedure would show the presence of an abnormal amount of non-volatile acid in the blood; it would not show the existence of actual *meionexy* *in vivo*. It would give precisely the same clinical information as would the Van Slyke test for blood bicarbonate unaccom-

panied by a measurement of carbon dioxide tension. When I made these studies the Van Slyke apparatus had not been invented. Some of my results are shown in table 2. They would seem to indicate that in the cases studied there was little if any relationship between dyspnoea and the intensity of the acidosis.

The discrepancy may be due in part to changes in the sensitivity of the respiratory center. It is getting progressively apparent that this may alter in disease, as Barr and Himwich found it does after exercise in normal persons, and therefore may be an important factor

TABLE 2

CASE NUMBER	CONDITION	OXYGEN SATURATION OF CO ₂ FREE BLOOD AT 17 MM. O ₂
		<i>per cent</i>
1	Normal control.....	63
2	Normal control.....	67
3	Normal control.....	68
4	Arteriosclerosis-uremia-Cheyne-Stokes breathing.....	10
4	After alkali. Still has Cheyne-Stokes breathing.....	61
5	Chronic nephritis, uremia. No dyspnoea.....	21
6	Chronic nephritis, uremia. No dyspnoea.....	57
7	Cardio-renal disease. Marked dyspnoea.....	44
7	On alkali. Less dyspnoea.....	52
8	Hemiplegia. Cheyne-Stokes breathing.....	52
9	Arteriosclerotic nephritis. Marked dyspnoea.....	50
10	Chronic nephritis, uremia. No dyspnoea.....	42
11	Lobar pneumonia.....	63
12	Pernicious anemia.....	70

in determining the inception of dyspnoea. As Fraser, Ross and Dreyer have put it,

It seems probable that the center may be affected in a great variety of ways, and that our knowledge of dyspnoea in diseased persons cannot be materially advanced until we take into consideration, not only the complicated conditions of the circulating blood, but also the condition of the respiratory center and the pathological changes that may occur in it, independent of oxygen supply, carbon dioxide tension, and hydrogen ion concentration.

The point that I should like chiefly to emphasize in regard to the relationship between acidosis and dyspnoea is that acidosis is a direct

producer of hyperpnoea, and that hyperpnoea will give rise to dyspnoea to a varying degree depending in part upon its intensity and in part upon those factors which determine the available supply of pulmonary ventilation. Since the acidosis of renal disease is more likely to be accompanied by circulatory or pulmonary defects than is that of diabetes, it is accordingly more likely to give rise to dyspnoea. The acidoses that produce no dyspnoea while the subject is at rest may do so during muscular work exactly as in hyperthyroidism.

IX. DYSPNOEA ASSOCIATED PRIMARILY WITH DIFFICULTY IN OXYGEN TRANSPORT

The usual stimuli to the respiratory center and through it the chief regulator of pulmonary ventilation, we have seen, is the reaction of the blood reaching the brain.¹⁰ We have also noted that under ordinary circumstances ventilation and blood flow are apparently more than ample to satisfy the requirements of oxygen transport. Nevertheless, it is obvious from a teleological viewpoint that one of the prime requisites in bodily economy must be adequate assurance of a free supply of oxygen. We are brought, therefore, to the question, how under abnormal circumstances does the organism respond to a threatened or impending oxygen shortage?

The effects of a diminished tension of oxygen in the inspired air have been widely studied. The researches of Haldane and his associates throw a bright light upon the rôle played by lack of oxygen in the regulation of breathing. Mountain sickness and the physiologic effects of living at high altitudes have been actively investigated by such expeditions as those of Barcroft to Monte Rosa and Teneriffe, and the Anglo-American expeditions to Pike's Peak and to the Andes. The effect of altitude upon the aviator during and since the War has been the subject of careful study by Schneider.

It is obvious from the oxygen dissociation curve of blood that as the oxygen tension of the alveolar air falls a time will come when the blood will be but incompletely saturated in the lungs. It is also clear that when the steep part of the curve is reached this failure to become

¹⁰ A still more modern theory is that the reactions of the tissues of the center itself is the true stimulus. Cf. R. Gesell.

saturated as the tension drops will rapidly assume large proportions, and that unless compensatory factors come into play, the point would soon be reached at which the organism would die of asphyxia.

An increase in pulmonary ventilation, provided there is no alteration in metabolism, will reduce the tension of carbon dioxide in alveolar air and blood. By the same token it is clear that within certain limits it will likewise raise that of oxygen. Therefore in the presence of a decreased tension of oxygen in the inspired air, the organism could compensate to a certain extent by hyperpnoea.

That such a reaction actually occurs, or in other words, that oxygen lack per se may stimulate the respiratory center either directly or indirectly has been proved by the studies of Haldane. In brief, the findings were that as the percentage of oxygen in the air breathed or the barometric pressure is reduced, no effects appear until oxygen tension has fallen about one third, and beyond that point hyperpnoea makes its appearance, at first in the form of a combined increase in rate and depth of respiration, as in the reaction to carbon dioxide. This first effect Haldane believes due to the lowering of the threshold of the respiratory center to its normal stimulus by the lowered oxygen tension. With a reduction in oxygen tension periodic or Cheyne-Stokes breathing makes its appearance, and finally with great reduction in oxygen tension the breathing becomes very rapid and correspondingly shallow, entirely different from carbon dioxide hyperpnoea. The periodic breathing and the rapid shallow breathing he regards as the direct effects of oxygen want, per se, on the respiratory center, and the latter when it assumes an extreme type as evidence likewise of fatigue or exhaustion of the center.

The evidence for the theory that periodic breathing is the result of oxygen want is very conclusive. The classical work on the subject is that of Douglas and Haldane. These workers found that they could produce typical Cheyne-Stokes breathing in themselves either by doing forced breathing or by enlarging their dead space by breathing through a long tube. After the apnoea which follows forced breathing they found that when breathing was resumed, usually at first for a period of about three minutes, it was of the Cheyne-Stokes type. Douglas' and Haldane's observations of the alveolar air showed that during the apnoea the oxygen tension rapidly fell to

about 32 mm., at which point breathing was resumed. During this same period the carbon dioxide tension rose rapidly at first, then slowly, but when breathing was resumed it was still some 3 mm. short of normal. The first period of breathing, induced they believed through oxygen want, rapidly raised the alveolar oxygen tension to about 75 mm., and at the same time lowered that of carbon dioxide some 2 mm. Apnoea began again and lasted until the oxygen tension fell to about 42 mm. Then another oxygen want period of breathing ensued and the cycle continued, oxygen and carbon dioxide tension both gradually rising until when the latter had reached its normal level continuous breathing was resumed. When the experiment was repeated, but with a rich oxygen mixture, no periodic breathing appeared, the same was true in breathing through a long tube. Moreover, it was found clinically by Pitt, Pembrey and Allen that clinical Cheyne-Stokes breathing could be abolished by the administration of oxygen or carbon dioxide, thus providing the normal stimulus, or by giving a gas mixture which contained less oxygen than air. This last they thought abolished periodicity by increasing the oxygen want and so providing a continuous instead of an intermittent stimulus.

The explanation offered by Haldane as to why breathing induced by oxygen want may be periodic while that induced normally by carbon dioxide is continuous is as follows: since there is a vast storage capacity for carbon dioxide in the body tissues and fluids, but practically none for oxygen, the action and cessation of stimulus occur more sharply in oxygen want than when carbon dioxide is the stimulus. Haldane says,

Thus the respiratory governor becomes too sensitive, just like the governor of an air engine when there is no flywheel or an insufficient load, and begins to "hunt," the oxygen supply to the center becoming, when the anoxemia is not too considerable, alternately so small as to cause urgent breathing and so great as, in the absence of carbon dioxide which has been washed out, to produce apnoea.

To plot a general curve for the reaction to oxygen want, such as that shown in figure 7 for carbon dioxide, is quite impossible. In the first place subjects would show marked individual differences and the

same subject would vary on different occasions. It also would be difficult to distinguish between effects due to lowering of the carbon dioxide threshold and those due directly to oxygen want stimulation. Nevertheless, in this connection the effect upon total ventilation and upon the tension of the alveolar gases during a progressive fall in the partial pressure of oxygen in the air breathed will be illuminating.

In figure 15 I have redrawn a curve of Schneider's which was obtained in testing men for the aviation service of the United States army. The subject breathed through sodium hydrate into a closed system. The carbon dioxide he gave off was all absorbed by the so-

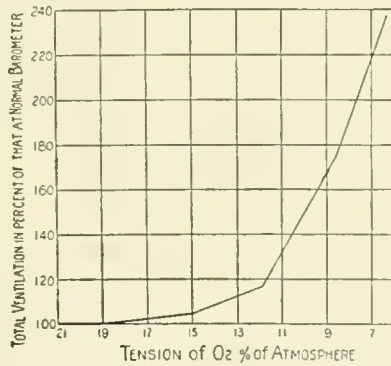


FIG. 15. VENTILATION REACTION TO OXYGEN WANT

The total ventilation, as the oxygen tension of the inspired air is reduced, is expressed in per cent of that which the subject had when breathing pure air at the barometric pressure of sea level. From data published by Schneider.

dium hydrate but the oxygen in the mixture gradually fell as his metabolism used it up. It will be noted that the total ventilation was doubled when the partial pressure of oxygen had fallen to 8 per cent of an atmosphere. It will also be noted that practically no reaction occurred until the oxygen of the inspired air had fallen to 15 per cent. Aside from this delay in the start, the curve perhaps bears some resemblance to the carbon dioxide curve (fig. 7). The particular man for whom this curve was obtained was "Class AA" an "extra good man." Schneider, unlike Haldane, found that the chief change in breathing even at oxygen tensions as low as 8.5 and 6 per cent of an atmosphere

was chiefly in amplitude rather than rate. Some individuals were found who showed little or no ventilation increase with reduction in oxygen, or who started to react and then with further "increase in altitude" suddenly began to breathe less. Such individuals were likely to faint and were quite unfit for flying at high altitudes. The reaction was further found to vary in the same individual at different times. When "stale," for example, the reaction was less than when in good flying form.

The condition in which the tension of oxygen and therefore the saturation of the blood is lower than normal is called anoxemia. It may result from internal (pathologic) causes as well as from those of external environment. Anoxemia should be regarded as evidence of difficult, not of incomplete, transport of oxygen. Such observation of the metabolism of anoxemic persons as can be found in the literature points not only toward no reduction in total oxygen absorption, but indeed sometimes toward an increased one. When the supply of oxygen to tissue cells becomes less than their metabolism calls for, it is to be presumed that death takes place from asphyxia. Anoxemia should be looked upon as denoting not partial asphyxia, but rather impending asphyxia. As long as life is to continue the required oxygen must be got in, but it may be only with great difficulty or after profound readjustments.

Barcroft has divided anoxemia into three main types which perhaps can best be illustrated diagrammatically. The amount of oxygen removed from the blood under normal circumstances and the coefficient of utilization of the oxygen carrying power have been discussed in previous sections. Under abnormal circumstances Barcroft points out anoxemia may result, (1) when there is deficient saturation in the lungs, anoxic anoxemia; (2) when there is an abnormally great reduction in the capillaries, stagnant anoxemia, and (3) when the blood is deficient in oxygen carrying power, anemic anoxemia. The actual happenings in these three types are shown in figure 16 in which columns representing arterial and venous blood are superimposed on the dissociation curve. The diagrams show both the concentration and percentage saturation with oxygen of arterial and venous blood, together with the tension of oxygen.

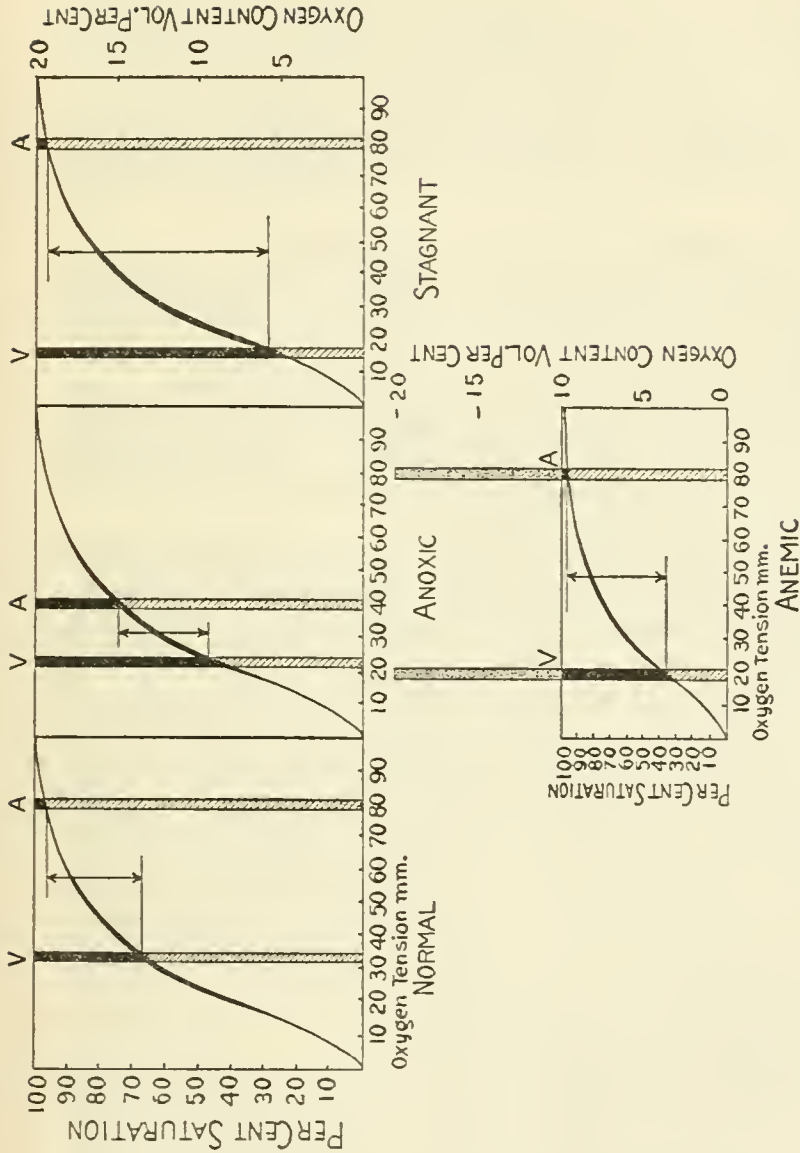


FIG. 16. DIAGRAM ILLUSTRATING TYPES OF ANOXEMIA

Columns representing arterial blood (A) and venous blood (V) are superimposed upon the oxygen dissociation curve. The black portion of the column represents reduced hemoglobin and the shaded portion, oxygenated hemoglobin. In the case of anemic anoxemia the dotted portion of the column represents hemoglobin that is either lost, as in true anemia, or unfit for oxygen transport, as in carbon monoxide poisoning. The perpendicular arrows denote the volume of oxygen delivered to the tissues from a unit of blood.

For our purposes the important thing to remember is that the anoxic type is due to faulty oxygenation in the lungs, and is therefore primarily due either to disease of the lungs as in pneumonia, emphysema or edema, or to deficient entry of air into them as in bronchial asthma and capillary bronchitis, or to bronchial, tracheal or laryngeal obstruction. The anoxemia of deficient oxygen tension in the inspired air of course is of this type also (mountain and aviation sickness).

Stagnant anoxemia is primarily due to decreased blood flow as in heart disease, massive hemorrhage or shock. The blood passing through the lungs is aerated properly, the arterial blood shows no anoxemia, but as the blood flow is too slow in relation to metabolic call for oxygen, there is an unusually large proportion of that gas removed in the capillaries with the result that the venous blood is more completely reduced than normally. It is an anoxemia, demonstrable by blood gas analysis, in the venous blood only. The anemic type we will discuss a little later.

The presence of anoxemia is often but not always made manifest by cyanosis, and its relation to anoxemia and the question of the mean capillary oxygen tension has been thoroughly discussed by Lundsgaard and Van Slyke in a recent number of this journal. The chief points made by them which it will be important for us to remember here are, first, that although cyanosis denotes anoxemia, anoxemia may exist without cyanosis. For example, Lundsgaard has shown that a certain concentration of hemoglobin is necessary before cyanosis becomes distinguishable. In severe anemia even with marked anoxemia no blue color would be apparent. Secondly, although cyanosis denotes anoxemia, it is difficult to interpret intensity of anoxemia from intensity of cyanosis because of a variety of factors which affect the latter, such as thickness of and amount of pigment in the skin, color of the blood plasma and variations in the number, width and length of blood filled capillaries in a given skin area.

The matter of cyanosis concerns us here directly only in so far as it is a manifestation of anoxemia, and that because anoxemia has been shown to be per se a cause of hyperpnoea, and therefore in accordance with our previous argument, a potential cause of dyspnoea. The rôle played by anoxemia in the production of dyspnoea is no less important

but far harder to evaluate than that of the factors discussed in earlier sections. The amount of hyperpnoea for a given rise in metabolic rate or change in blood bicarbonate level can be predicted tolerably accurately. Schneider's work shows that that of anoxemia cannot. At present we must content ourselves with the knowledge that anoxemia is capable of producing dyspnoea either of itself or more often, as we shall see later, in conjunction with other factors.

The most important thing to appreciate is that the effects of anoxemia, if of marked degree, may be of a serious nature and that its recognition is important. The symptoms produced by it are varied. They have been observed and described in both the sickness of the mountaineer and of the aviator as well as under experimental conditions. In the patient, they are usually combined and possibly masked by other symptoms of the underlying disease and are therefore doubly difficult to interpret.

Even when the pathognomonic signs, as I think we may call them, of anoxemia, cyanosis or periodic breathing are present, it is difficult to estimate the intensity of the anoxemia or its relative harmfulness. The type of anoxemia and likewise its duration make important differences both in its power to do damage and in its tendency to produce hyperpnoea and dyspnoea.

Haldane's work has shown that it is the tension of dissolved oxygen which is important for tissue respiration and which, when it falls below a certain point, acts as a stimulus to the respiratory center. Of course the oxygen content is important as well, for with the tissues taking up oxygen at a constant rate, the less the load of oxygen, the greater will be the withdrawal from each unit of blood, and consequently the lower the mean tension.

Of the anoxic type of anoxemia I shall have something further to say in the section on pneumonia, and of the stagnant, in that on cardiac dyspnoea. In concluding the present section I should like to consider briefly the type of anoxemia due to abnormality of the blood itself. From the last diagram in figure 16 it is clear that in anemia there is a reduced amount of oxygen per unit of blood. Nevertheless the oxygen tension and saturation of the arterial blood is normal. There is no impediment to oxygenation in the lungs. Since the call for oxygen by the tissues is no less (in fact in primary anemia

and the anemia accompanying leukemia it may be increased) one of two things (or a combination of two things) must happen, either a relatively larger quantity of oxygen must be removed from a unit of blood as it passes through the tissue capillaries, thus giving an anoxemia in the mean capillary and venous blood, or the rate of blood flow must be increased in proportion as the oxygen carrying capacity of the blood is reduced. These two principles are illustrated in figure 17.

Should the first alternative occur there would be no difficulty in gas transport during rest, and I believe it is a matter of common experience that even severe anemia (except that due to sudden massive hemorrhage, and in that case the anoxemia is rather of the stagnant than the anemic variety) gives rise to no hyperpnoea or dyspnoea so long as the patient remains quiet. The normal person utilizing perhaps but 25 to 30 per cent of his oxygen carrying capacity has a good portion of the remainder as a reserve which upon exertion lies between him and asphyxia. The anemic patient, unless his blood flow is accelerated, sacrifices a certain part of this factor of safety, the degree of sacrifice depending on the degree of anemia. Under these circumstances the performance of muscular work might produce anoxemia of marked degree and hyperpnoea and perhaps dyspnoea would result.

If the second alternative occurred, that is to say compensatory acceleration of blood flow, then there might be no anoxemia, not even venous, but the heart would be required to do continually an abnormal amount of work, for the work of the heart depends upon the amount of blood it pumps in a unit of time and the pressure against which it is pumped. The performance of muscular work under these circumstances would call for a still greater circulation rate and hyperpnoea and dyspnoea might result from insufficient blood flow just as a little later we shall see they do in cardiac disease.

Which of these alternative adjustments actually takes place in anemia remains to be proved. Haldane and his school believe the latter. Lundsgaard inclines to the former. Haldane based his conclusions on blood flow determinations made on the Fick principle, Lundsgaard on the arterial and peripheral venous blood. These principles have been discussed earlier. Each is open to question, the

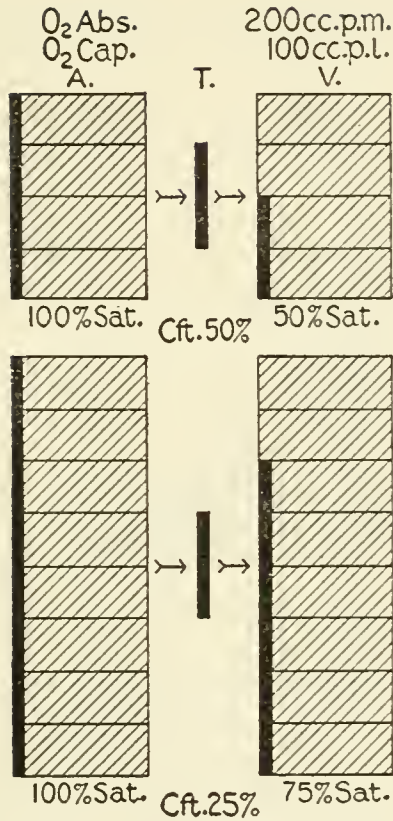


FIG. 17. SCHEMA OF THE BLOOD FLOW IN ANEMIA

Compare figure 9. The oxygen absorption is assumed to be the same as that of the normal person, 200 cc. per minute. The oxygen capacity, because of the reduction in hemoglobin, is only 100 cc. per liter. There must be a variation, therefore, from the normal either in the coefficient of utilization of oxygen carrying power, as is shown in the upper portion of the diagram, or in the total blood flow as is shown in the lower. The first instance shows what would happen if 4 liters of blood flowed per minute; 200 cc. would be left in the tissues and the venous blood would have a saturation of 50 per cent; the coefficient would be 50 per cent. The second instance shows a doubling in the blood flow, 8 liters per minute, with the result that the venous blood is 75 per cent saturated and the coefficient is 25 per cent.

truth may well lie between the results obtained by the two. If the coefficient of utilization of the oxygen carrying capacity could be accurately determined the problem would be solved, but to date it has not been.

A series of coefficients in normal persons and in patients with polycythemia and anemia based on arterial and peripheral venous

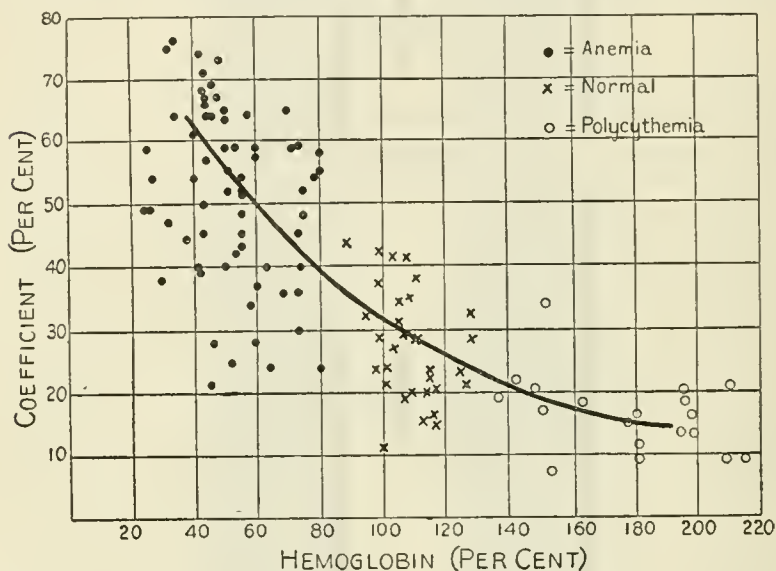


FIG. 18. THE COEFFICIENT OF UTILIZATION OF OXYGEN CARRYING CAPACITY WITH VARYING CONCENTRATIONS OF HEMOGLOBIN

The ordinate shows the coefficient; the abscissa the percentage of hemoglobin. The points were determined by personal observation or collected from the literature by Bock and Means. Although the points show a considerable individual scattering, there is, nevertheless, a definite inverse ratio between the coefficient and the hemoglobin concentration.

blood was collected by Dr. A. V. Bock and myself in 1920 from personal observations and from the literature. They are shown in figure 18. Because of the variability of arm vein blood they can be taken as suggestive evidence only. It may be said, however, that much as does the oxygen saturation of arm vein blood vary, normally it does not vary to the extent shown by these coefficients obtained in blood diseases. Furthermore the tendency for the coefficient to be

consistently higher as the hemoglobin falls and lower as it rises above normal makes it seem unlikely that in anemia decreased oxygen carrying power is completely compensated by increased blood flow.

In those conditions of the blood in which a portion of the hemoglobin is useless for oxygen transport, such as carbon monoxide poisoning, nitro-benzol poisoning and methemoglobinemia there probably is anoxemia as in ordinary anemia. These conditions, however, are often acute while anemias, except that due to sudden hemorrhage, are for the most part chronic. The general reaction to anoxemia in the former, therefore, may be somewhat different for it has been shown that acute oxygen want gives rise to a different picture than chronic.

X. DYSPNOEA ASSOCIATED PRIMARILY WITH MECHANICAL AND NERVOUS HINDRANCES TO THE RESPIRATORY MOVEMENTS

Under this heading I shall consider those factors which mechanically, or through nervous channels, either limit the total pumping capacity of the pulmonary bellows or which force upon it an inefficient type of respiration.

It is apparent from what has been said earlier that the point at which hyperpnoea gives rise to dyspnoea will bear some relation to vital capacity. The ventilatory capabilities of persons with reduced vital capacity are obviously less than those with normal ones. As the volume per breath approaches that of the vital capacity the breathing tends to become distressing.

There are many causes for reduced vital capacity. Rabinowitch's findings in exophthalmic goiter have been discussed. The reduced vital capacity of heart disease and pneumonia will be considered in the sections which follow. Other causes for reduction are often found, such as hydro- or pneumothorax, mediastinal or thoracic tumors. The effects of these various causes are similar.

Pneumothorax presents a reasonably simple problem, a reduction in vital capacity essentially unaccompanied by other factors which might influence respiration. Balboni and I have made some observations upon the breathing in this condition. In the first place it was found that basal metabolism, alveolar carbon dioxide tension and rate and amplitude of breathing were all within normal limits during

rest. The vital capacity on the other hand, as would be expected, showed a diminution to the neighborhood of one-half of the normal. The reaction to carbon dioxide was observed in several cases, and the finding in brief was that the increase in total ventilation followed the normal curve (fig. 7) until the increase reached about 300 per cent at which point dyspnoea became marked and the observation had to be discontinued. It will be remembered that in the normal the dyspnoea point was reached at between 400 to 500 per cent. In other words these patients with pneumothorax presented an example of dyspnoea on exertion only, due entirely to a mechanical limitation in their ventilatory capacity. Within their capacity they react in a normal manner but their capacity is materially diminished. Should their rate of metabolism because, let us say, of the performance of muscular work exceed their ventilatory limitation, they would of course have a retention of carbon dioxide (carbonic acidosis) and very likely anoxemia. The onset of dyspnoea, however, forces them to cease work before any such point is reached.

Another condition in which a reduced vital capacity results of necessity from the existing anatomical abnormality is pulmonary emphysema. The state of affairs here is, however, somewhat more complicated than in pneumothorax. Besides the lowered vital capacity there is, as Hoover has shown, an increase in size of the dead space which he believed due to bronchiolar atony. We have seen that increased dead space is a primary cause for hyperpnoea because it necessitates a greater total ventilation for the accomplishment of a given alveolar ventilation. The emphysematous patient therefore has in his increased dead space a respiratory handicap constantly present which he must overcome, just as the thyrotoxic patient has in his increased rate of metabolism. Furthermore his lowered vital capacity diminishes his total ventilatory capability, thereby reducing the amount of effort that can be made without dyspnoea.

Emphysematous patients, however, in contrast to those with pneumothorax, present evidence of a certain degree of compensatory adjustment to these adverse circumstances. R. W. Scott for instance has made the interesting discovery that in emphysema there is an increased tolerance to carbon dioxide. That is to say, as his patients breathed increasing percentages of carbon dioxide their reaction

curves, instead of following the normal as did those with pneumothorax, were much flatter. For a given rise in the carbon dioxide concentration of the inspired air there was less percentage rise in total ventilation in emphysema than in normal persons. The explanation of this phenomenon lies in the supernormal level of blood alkali in these cases. Scott found, and others had found earlier, that the alveolar carbon dioxide tension was increased in emphysema. Scott also found that blood reaction was not abnormal. In other words the conditions of acid-base balance must be those as shown in the carbon dioxide diagram (fig. 12) by point 5 (high level curve with normal pH).

This state of affairs in emphysema to a certain extent amounts to a compensation for a ventilatory handicap, for it is obvious from the second law of breathing that it reduces the amount of ventilation necessary to eliminate a given quantity of carbon dioxide. This is far from an unmixed blessing. It does, it is true, diminish the labor of breathing, but at the same time reduces the margin of safety against oxygen want. If through high level of blood bicarbonate ventilation is decreased too much, anoxemia may result, and the compensatory mechanism while diminishing one evil may introduce another.

In point of experience Scott found that although his emphysematous patients could tolerate more carbon dioxide than normal persons, they also, when they did reach the breaking point, reached it far more acutely and distressfully than do normal persons. It may be inferred that their compensation at best is but an imperfect one, and that not only are they often constantly slightly anoxic, but that frequently they are in danger of severe anoxemia. Of course with emphysema there often is associated cardiac pathology. Under such circumstances we might have added all the factors of circulatory insufficiency as well.

The problem presented by bronchial asthma is an interesting one. In this condition we have an element that is probably unique, in the prolongation of the expiratory phase. The bronchiolar spasm doubtless throws some difficulty in the way both of the entrance to and exit of air from the lung alveoli, but this difficulty is more apparent in the expiratory phase because of the fact that expiration ordinarily is passive. In the asthmatic attack an active expiration is required

which greatly reduces the point at which the breathing becomes distressful. Furthermore the vital capacity is much reduced so that the asthmatic with lowered ventilatory capability encounters active resistance in both phases of the respiratory movements, and suffers therefore from intense dyspnoea. The ventilation is, very likely, often frankly insufficient in asthma, and anoxemia and cyanosis result. Spirographic tracings recently taken by S. G. Mudd in the medical clinic of the Massachusetts General Hospital show that the well known relief afforded by epinephrin during the asthmatic attack is accompanied by an increase in the volume per breath and in the total ventilation and vital capacity, and a shortening of the expiratory phase. The relaxation of the bronchiolar spasm relieves the obstruction to the free passage of air, increases the vital capacity thus removing the chief causes of the dyspnoea.

The likelihood of intrathoracic tumors producing dyspnoea seems to depend to a greater extent upon their position than upon their size. A tumor situated laterally would, to be sure, encroach upon the lung to a certain extent thus reducing vital capacity but reduction from such a cause would rarely be of a degree that would give rise to more than a slight ventilatory handicap. It is the centrally placed, that is mediastinal, tumors that produce the greatest embarrassment. This may be in part due to actual pressure narrowing of the trachea or bronchi, but perhaps in part also to interference with the proper action of the thorax or diaphragm. I recall particularly in this connection two patients.

One was a young man with acute lymphatic leukemia who suddenly developed a rapid and very distressing type of breathing. His total ventilation per minute was 8.7 liters and the volume per respiration 415 cc. A determination of the basal metabolism showed an increase of 50 per cent above normal. His carbon dioxide output therefore was increased 50 per cent and so approximately was his total ventilation, but this amount of increase per se should not have caused dyspnoea. His vital capacity on two occasions was only 1.1 liters. This is less than one-third of what it should have been. A slight hyperpnoea with a marked restriction in vital capacity accounts for the dyspnoea. In normal persons it will be remembered the volume per breath is about 12 per cent of the vital capacity, in this boy it was

38 per cent. At autopsy the lungs were clear, but in the mediastinum there was a hard mass of lymph nodes the size of a man's fist. This mass was the only anatomical cause that could be found for the low vital capacity.

The second patient was a man of seventy-two who had had very severe dyspnoea coming on quite acutely five weeks before he came under observation. One interesting feature of this dyspnoea was that it was worse when he sat up than when he lay flat, that is, the antithesis of orthopnoea. He also had cyanosis of the head and upper extremities which was most marked when he lay down. There was edema of the face and neck with supracardiac dullness; dilated veins about the insertion of the diaphragm and a moderate hydrothorax on the right. In other words he presented the picture of upper mediastinal pressure. At autopsy there was found a malignant lymphoma of the mediastinum pressing on the vessels to and from the heart. Of course there were several causes for dyspnoea in this man. An anoxemia hyperpnoea was probably one. Nevertheless it seems likely that the major portion of the dyspnoea was mechanically produced and the interesting thing for us to note now is that in this patient the volume of air per breath and the vital capacity were essentially identical, about 500 cc. The respiration rate was 35. Not only was there a rapid rate but each breath, though not larger than normal, was yet 100 per cent or thereabouts of the vital capacity instead of 12 per cent or so. Each breath required a great effort and the dyspnoea accordingly was extreme. Moreover this was true during complete rest. He had absolutely no power to increase ventilation further. Muscular exertion therefore was impossible.

Interference from any cause with the proper action of the diaphragm probably limits ventilatory power or at least lowers the dyspnoea point through the throwing of added work on the thorax, although Schroeder and Green have concluded from their experimental work that the diaphragm is not an essential muscle of respiration. Sudden paralysis of the phrenic nerves gives rise to more respiratory embarrassment than gradual. Abdominal distention, particularly when it comes on acutely, perhaps through an effect upon the diaphragm, seems capable of making the respiratory movements distressful. I have seen several patients who when the stomach or bowels were

distended with gas had a rapid shallow type of breathing, one might call it tachypnoea, which was distinctly unpleasant. The effect of abdominal distention will be discussed somewhat further in the section on pneumonia.

Under mechanical hindrances the last which I should like to mention is obstruction to the passage of air as from narrowing of the bronchi, trachea or upper air passages. The type of dyspnoea so produced is familiar enough to all and needs no comment here, the experimental results, however, of Davies, Haldane and Priestley are of interest in this connection. These investigators produced obstruction to the respiration of normal persons. It was found that the response to such resistance was a slowing and deepening of the breathing. They also found that when the resistance becomes excessive the respiratory center begins after a time to become fatigued and the breathing then becomes shallow and more frequent. This produces anoxemia which in turn aggravates the fatigue of the center and a vicious circle is established.

It is probable that purely nervous causes, other than paralysis of the motor nerves of the respiratory muscles, are capable of limiting ventilatory capacity and hence may be potential causes of dyspnoea. We have seen that while the chemical control of breathing determines the quantity of alveolar ventilation, the nervous control determines the type of breathing that shall accomplish the required ventilation. If this is true, it is quite conceivable that a disturbance in innervation might so interfere with normal expiration that a state of constant inflation might be established. Considerable evidence in favor of this view will be found in the work of White on shell-shocked soldiers and other psychoneurotics. Some of these men with no organic lesions of the heart or lungs suffered from all the subjective symptoms of cardiac failure, dyspnoea, palpitation and precordial pain. The striking thing is that while objectively there was no organic heart lesion, nevertheless the vital capacity was often reduced, the degree of reduction varying with the degree of nervousness. It is hard to attribute this limitation of vital capacity in these soldiers to any but nervous causes, but even so it handicaps as one organically produced.

In summary it may be said that any factor which reduces vital capacity, be it functional or anatomic, will lower the dyspnoea point,

and that any factor which increases the dead space will increase total ventilation and thus be a primary cause of hyperpnoea and therefore potentially also of dyspnoea.

XI. CARDIAC DYSPNOEA

The cardinal symptom of the failing heart is dyspnoea, especially the kind that is definitely related to exertion, but also the paroxysmal variety, so-called cardiac asthma, as well as the related manifestations, orthopnoea and periodic breathing.

In attempting an analysis of the causes of cardiac dyspnoea we shall do well to follow our original scheme, starting therefore with metabolic respiratory demand. The work of Peabody, Meyer and DuBois shows that there is often an increased metabolism. These investigators studied the heat production and elimination in cases of cardiac and renal disease. They say:

Patients with compensated cardiac lesions or with mild nephritis showed no increase in the metabolism. Of twelve patients with dyspnoea, nine showed a distinct rise in metabolism, and in five of these the increase was from 25 per cent to 50 per cent above the average normal. Two out of the five gave evidence of marked acidosis in the low content of carbon dioxide in the alveolar air. In two others, whose metabolism was just as high, there was no significant depression of the alveolar carbon dioxide.

We may say then that from the point of view of demand for ventilation, the patient with cardiac insufficiency may have anywhere from no increase to an increase of 25 to 50 per cent. This per se would not be sufficient to cause dyspnoea during rest, but in the way previously described would play its part in the lowering of the point at which dyspnoea would appear on exertion.

The ability of the blood itself to transport gases is probably adequate in cardiac disease. Such changes as have been observed in the oxygen or carbon dioxide combining power are undoubtedly secondary to impaired circulation. Increased oxygen capacity is frequently met in the familiar erythrocytosis of congenital and of chronic acquired heart disease, especially when associated with marked cyanosis, and may reasonably be regarded as a compensatory phenomenon. Depressions in carbon dioxide capacity may be associated with anoxemia

in the cardiac patient just as they are in the normal person while residing at a high altitude, but there again we are dealing with an effect rather than a cause. Factors demanding hyperpnoea of a primary nature, that is inherent in the blood as in true acidosis, such as either the metabolic variety of diabetes or the retention variety of nephritis, probably do not exist in uncomplicated heart disease. They would occur only when renal disease is present as well.

The fundamental fault responsible for cardiac dyspnoea is obviously to be found not in the nature of the blood but in the rate at which it is pumped; in the heart itself. Subnormal circulation rate, which undoubtedly exists in many if not all patients with heart disease, may be brought about in a variety of ways; leaking or obstructed valves, overworked or weakened heart muscle or by congenital lesions with communication between the two sides of the heart.

Into the varieties of heart disease we need not go. The factors leading to dyspnoea are common to all. The important point is that the heart either because of increased work, fatigue or degeneration is unable to maintain an adequate rate of blood flow. Increased work may be due to valvular defects, to adhesive pericarditis, or to hypertension or hyperthyroidism, while degeneration may be due to infectious or toxic myocarditis, or to arteriosclerosis. The results as far as dyspnoea is concerned are not so very different.

Let us consider therefore the effects which decreased minute volume of blood flow may have upon gas transport and hence upon external respiration. We may for convenience separate these effects into those upon the carriage of each of the two respiratory gases, recognizing, however, that the two are inseparably interrelated.

Oxygen transport, being the more conspicuous since difficulty with it gives rise to the cyanosis that we attribute so confidently to disease of the heart, is perhaps best to discuss first. The types of anoxemia have been described earlier. The stagnant is that primarily due to slow blood flow; the anoxic or arterial that primarily due to deficient aëration in the lungs. Both may occur in cardiac failure, but the former may also occur in heart disease before failure has set in.

Certain cardiac lesions more than others materially interfere with the heart's output, stenosis and insufficiency of valves, especially mitral stenosis, also those congenital lesions in which there is a left

to right shunt. In cases of these types reduced blood flow is probably a constant affair. It is a natural consequence of the lesion and does not of itself indicate failure. Of course in such cases even when compensation is good, there is an extra burden on the heart and therefore on the respiratory organs and hence a lowering of the dyspnoea point, that is, dyspnoea on exertion of a grade that would cause none in a healthy person. In all types of heart disease there probably is decreased blood flow when failure sets in.

The methods of determining blood flow in man together with the results obtained and the sources of error have been discussed in section V. Observations on patients with heart disease are scant. In 1915, Newburgh and I published figures for the rate of flow determined by the Krogh-Lindhard method during rest and work in a normal subject (J. H. M.) and in a man with chronic rheumatic heart disease (C. L.). This patient had double lesions of the aortic and mitral valves but was well compensated and had only slight cyanosis.

In figure 19 are shown the curves for oxygen absorption (metabolism), total pulmonary ventilation, blood flow and pulse rate for the normal subject (J. H. M.) at rest and with increasing amounts of work. It will be noted that blood flow, metabolism and ventilation show an even and similar rise as work increases. The pulse curve lags behind; it is reconcilable with the blood flow curve only on the theory that output per systole increases with work.

The curves for the cardiac patient (C. L.) are given in figure 20. The resting values for this subject by comparison with table 1 will be seen to be quite like those for normal persons. They were (average) blood flow 4.6 liters per minute, pulse 72, volume per systole 64 cc., coefficient of utilization of oxygen carrying power 39 per cent. During work it will be noted there is a sharper rise in metabolism (oxygen absorption) than in J. H. M. This would indicate that C. L. was a less efficient machine. The blood flow increase, while not unlike J. H. M's, does not keep pace with metabolism as did J. H. M's. This of necessity means a greater unsaturation of venous blood in the cardiac than in the normal subject. The coefficient for instance in J. H. M. rose from the resting value of 40 per cent to a maximum of 47 per cent during work. C. L's rose from a resting value of 39 per cent to a maximum of 55 per cent during work.

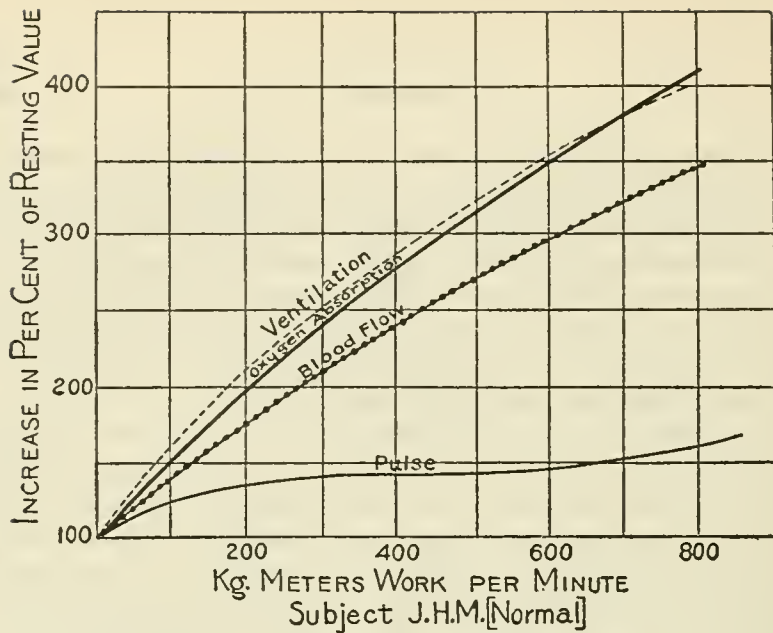


FIG. 19. THE BLOOD FLOW, OXYGEN ABSORPTION, PULMONARY VENTILATION AND PULSE REACTION TO WORK IN THE NORMAL SUBJECT (J. H. M.)

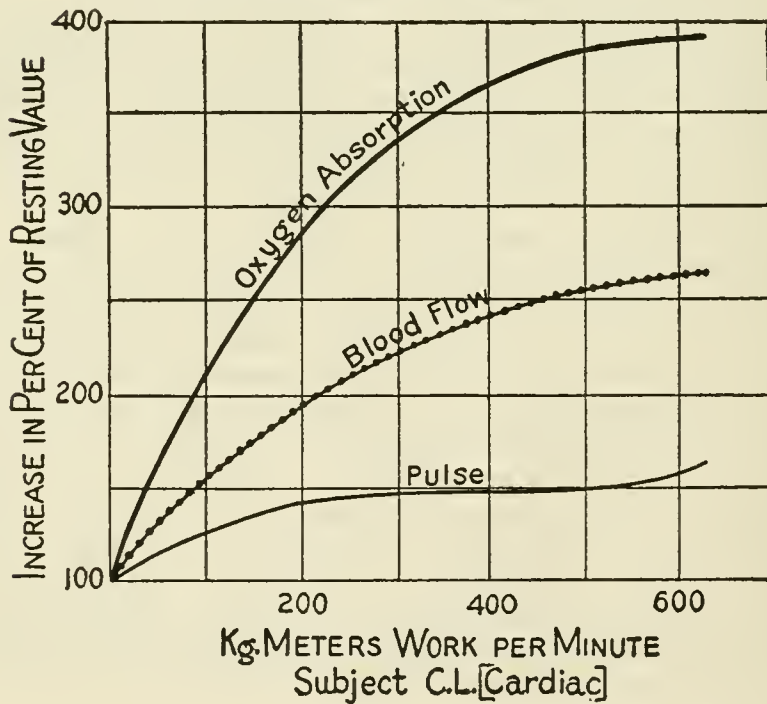


FIG. 20. THE BLOOD FLOW, OXYGEN ABSORPTION, AND PULSE REACTION TO WORK IN A CARDIAC SUBJECT (C. L.)

The gases of both arterial and venous blood in cardiac disease have received considerable attention in the last decade. The whole subject with special reference to cyanosis has been reviewed and discussed in a recent monograph of this series by Lundsgaard and Van Slyke. In general the evidence points toward stagnant anoxemia of varying grades due to slow blood flow, particularly in mitral stenosis (Means and Newburgh; Lundsgaard; Meakins, Dautrebande and Fetter).

Arterial or anoxic anoxemia also may occur in heart disease, but only when there is failure (Harrop). It is due to defective aëration in the lungs, perhaps from simple cardiac edema or to complications. Harrop for instance, in many of his patients with cardiac decompensation found that the arterial blood was only 85 to 90 per cent saturated instead of 95 per cent as in the healthy subject. His lowest finding of 81.4 per cent was in a patient with emphysema, chronic bronchitis and chronic myocarditis.

In congenital heart disease the arterial blood may be profoundly anoxic but here the difficulty lies not in the lungs (as is otherwise the case in this type of anoxemia) but in a right to left shunt in the heart itself. The anoxemia of such cases is unique in this respect. The most extreme grades of either arterial or venous anoxemia ever observed are found in these congenital cases. Such a patient has recently been studied at the Massachusetts General Hospital by Bock, Field and Henderson. Their findings will soon be published. According to their calculations the pressure head of oxygen available in this patient's tissue capillaries was about what a normal man's would be were he dwelling at the summit of Mount Everest.

As retarded blood flow brings about an undue unloading of the blood's oxygen in the tissues, so does it likewise lead to an excessive loading of carbon dioxide. This excessive load, however, can be blown off in the lungs by hyperpnoea. Pearce has emphasized this phase of the problem quite clearly and shown how it is possible for insufficient blood flow to be compensated, so far as carbon dioxide is concerned, by supraventilation. I have already touched in section V upon the relation between the minute volume of blood flow and ventilation in the normal. So far as we can judge they are of somewhere near the same order of magnitude.

In figure 21 I have attempted, following Pearce's conceptions, to represent diagrammatically the relationships which may be expected to exist in heart disease. On the left is shown what is believed to take place in healthy persons; a progressive and parallel rise in both factors until the point is reached as indicated by the dotted line where cardiac output can no longer keep pace with metabolic demand. At

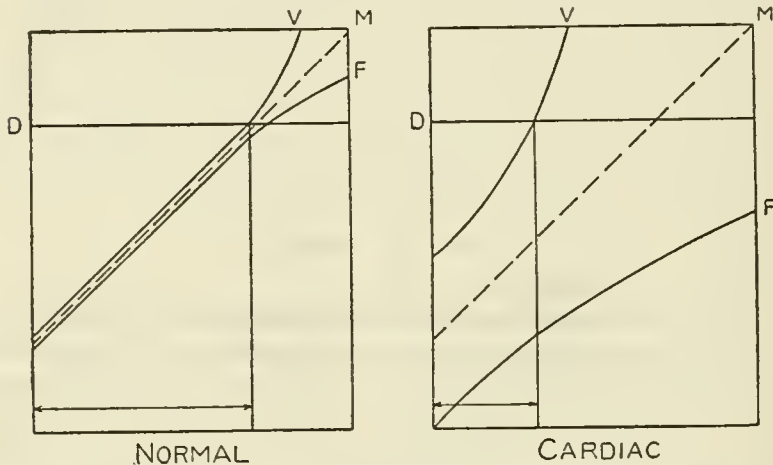


FIG. 21. SCHEMA SHOWING CHARACTER OF THE BLOOD FLOW AND VENTILATION REACTION TO WORK IN THE NORMAL AND CARDIAC SUBJECT

The ordinate denotes percental increase above resting value; the abscissa, work performed.

In both subjects the metabolism (M) increases progressively as work increases. In the normal, ventilation (V) and blood flow (F) show a like increase to a certain point. Beyond that F becomes less steep than M , and to overcome this V becomes steeper (superventilation). In the cardiac subject F is subnormal even during rest, V is therefore supernormal. When work begins F fails to keep pace with M earlier in the process, and superventilation as shown by the course of V begins sooner.

The horizontal line (D) represents the amount of hyperpnoea necessary to give dyspnoea. It will be seen that V reaches D at a much lower grade of work in the cardiac than in the normal.

this point superventilation comes into play to compensate for the insufficient blood flow. At the right is shown what may be expected to happen in cardiac patients.

In the first place the resting blood flow in heart disease may be slower than normal; therefore ventilation, even at rest, must be above normal. As metabolism rises with work, because of weakness either

relative or absolute of the heart muscle, blood flow begins to lag behind metabolism earlier than in the healthy subject. Superventilation therefore sets in earlier, and if we let the horizontal line D represent the level of ventilation at which dyspnoea appears, then it is obvious that the metabolic increment M possible to the cardiac patient is much smaller than that of the normal man.

An infinite number of diagrams of this sort might be made to indicate quantitatively the conditions existing in actual individual patients, for in no two probably are the factors precisely alike. The two qualitative diagrams that I have shown, however, may make clear the general character of the variations in heart disease and the nature of the task with which the cardio-respiratory mechanism is confronted.

The observations of Peabody and Sturgis seem to bear out the theories of Pearce. In patients with compensated valvular heart disease standing still, the minute volume of the ventilation was found to be greater, and the breathing faster and more shallow, than in healthy persons under the same conditions. During work of a mild grade the increase in ventilation was relatively the same in the normal and cardiac subjects studied, but absolutely greater in the latter. After exercise stopped the dyspnoea was found to continue longer in the cardiac subjects. It was concluded that the explanation for this lay in a delayed elimination of carbon dioxide due to inadequate blood flow.

In brief, then, we may assume that in cardiac disease there may through slow blood flow be delay both in getting oxygen in and carbon dioxide out. The latter may to a certain extent be compensated by superventilation. Superventilation has but little effect on anoxemia, none on the stagnant type, possibly a slight relief of the anoxic type through an improved tension of oxygen in the alveolar air. This phenomenon is seen in persons living at high altitudes. We should bear in mind that anoxemia per se is believed to be a direct cause of hyperpnoea.

The type of breathing in heart disease, as has been said, is often rapid and shallow. This is an uneconomical type for, as has been pointed out by Edsall, a relatively greater amount of total air pumping has to be done to accomplish a given ventilation of the alveoli than

with a slower and deeper type. Haldane, Meakins and Priestley furthermore believe that rapid shallow breathing tends to produce anoxemia; this being due, they think, to an incomplete expansion of the lungs and the passage of a portion of the blood through lung areas where it is incompletely oxidized. In these areas the carbon dioxide would be incompletely eliminated, therefore superventilation of the clear portions would be called for. In other words, this respiratory type of the cardiac patient's, as well as his slow blood flow, may in several possible ways be expected to throw an extra ventilatory burden upon him.

The reason for the shallow type of breathing lies chiefly in a restriction in the vital capacity, although according to the theories of the Oxford School it may in part be the results of anoxemia, for not only, so Haldane, Meakins and Priestley tell us, does rapid shallow breathing produce anoxemia but the hyperpnoea induced by anoxemia often is of a rapid shallow type. Thus a vicious circle is established. This, however, is all pretty theoretical, whereas the restriction in vital capacity is a well established fact.

That such restriction nearly always occurs in congestive failure has been shown by Siebeck, Bittorf and Forschbach, and by Peabody and Wentworth. Sometimes the capacity is as low as one liter. The midposition is apt to be depressed sometimes to such an extent that there is little or no reserve air. The complemental air is generally less reduced than the reserve. Peabody and Wentworth followed a number of cases through the period of failure into that of compensation and found that the vital capacity gradually increased. The amount of restriction in vital capacity and the amount of dyspnoea usually varied together, and the relative size of the vital capacity furnished a good index of any given patient's cardiac reserve.

The reason for the reduction in vital capacity in heart disease seems to be, according to the experimental results of Drinker, Peabody and Blumgart, the loss of elasticity of the lungs which results from congestion. At least this is an important factor. The enlargement of the heart and the extra blood in the pulmonary circuit would encroach somewhat on the available air space. Experimentally it was shown that when the pulmonary veins were partially occluded and the pressure in the pulmonary circuit thereby increased, the capacity

of the chest for containing air varied almost inversely with the pulmonary blood pressure.

In a recent paper Sturgis, Peabody, Hall and Fremont-Smith have studied the maximum minute volume of ventilation possible to normal man. They found that 35 per minute was about the highest rate and one-third the vital about the highest volume per breath that could be maintained for any appreciable length of time. The maximum possible ventilation for any subject therefore might be predicted according to the formula

$$\text{Maximum minute volume} = \frac{\text{vital capacity}}{3} \times 35$$

In other words, the vital capacity, they believe, is a good index of the respiratory capabilities of the cardiac patient, and hence of his ability to exert himself.

In summary we may then say that the amount of dyspnoea of any individual with heart disease will depend chiefly on the rate of his circulation and upon the vital capacity of his lungs. Both of these in turn are dependent upon the state of the heart muscle and the magnitude of the obstacles it has to overcome. The relative importance of these factors will vary from patient to patient and with the degree of failure or compensation and the type of lesion. Here we can only indicate certain underlying principles. It should be clear, however, why dyspnoea may occur on exertion in heart disease even when the myocardium is entirely adequate. As failure sets in certain complicating factors make their appearance, edema of the lungs for example, adding perhaps an anoxic to an already existing stagnant anoxemia, or renal insufficiency producing acidosis and hence a further call for hyperpnoea. All these increase the respiratory task and therefore the degree of embarrassment.

Before concluding this section a word on certain special forms of dyspnoea seen in cardiac patients is in order. I refer to cardiac asthma, Cheyne-Stokes breathing, and orthopnoea.

Cardiac asthma of course is simply a descriptive term applied to sudden attacks of dyspnoea of a sort resembling true bronchial asthma but occurring in chronic heart disease. Oftentimes there is true bronchiolar spasm with relief from adrenalin. Other attacks

simulating asthma may be due to sudden pulmonary edema, this variety being common in arteriosclerotic or in hypertensive heart disease. It is obvious that sudden edema of the lungs may cause very urgent dyspnoea both by its interference with gas exchange in the alveoli and by actual encroachment on available air space within the chest.

Cheyne-Stokes breathing Haldane believes always is a manifestation of oxygen want. This may be true, but the relationship is by no means a simple one. The state of excitability of the respiratory center is an important factor, and is doubtless subject to many influences. So simple a matter as the degree of sleepiness or wakefulness may determine whether or not the breathing of a given patient at a given moment will be periodic. Haldane is doubtless right in supposing that periodicity denotes imperfect regulation; the analogy with an engine governor is very apt. But we well know that anoxemia only occasionally is accompanied by periodicity. A variety of circumstances have to be just so before periodicity occurs, somewhat as in the case of a motor car which develops a certain rattle at 35 miles but not at 33 or 37.

For orthopnoea too Haldane offers an explanation. The fan-like expansion of the lung which in shallow breathing is thought to be incomplete, thus giving rise to anoxemia, is believed to be even more incomplete in the recumbent position than in the upright. To the normal person this matters but little; to the dyspnoeic cardiac patient on the other hand, already anoxemic while upright, it may amount to so much that breathing becomes well nigh impossible while lying flat. The recent work of Christie and Beams who have shown that orthopnoea occurs only when the vital capacity is reduced bears this out. These authors find that in 80 per cent of normal persons the vital capacity is less in the recumbent than in the upright position. If the original loss from disease is great and if a patient suffers an additional loss on lying down dyspnoea is necessarily intensified, that is to say, there is orthopnoea.

The effect upon the breathing in heart disease of such complications as emphysema, bronchitis or hydrothorax will be apparent from what has been said in section X; the effect of pneumonia will be discussed in section XII.

XII. THE DYSPNOEA OF PNEUMONIA

I have spoken in section II of DuBois' discovery that there is a rise of about 7.2 points in the basal metabolic rate for every degree Fahrenheit of fever. From this it will be apparent that the pneumonia patient with five degrees of fever will have to respire sufficiently to accommodate a gas exchange which is some 36 per cent higher than it would be in health.

The blood itself as a transporter of gases is probably not particularly abnormal in pneumonia. I can find no convincing evidence, for instance, that the oxygen combining power is materially altered. Studies of the carbon dioxide diagram (Bock, Means and Woodwell) (Barach, Means and Woodwell) have shown some tendency for the curve level to be slightly depressed and the pH to be altered slightly in the direction of acidity. After the crisis both curve level and pH become normal.

Why the arterial point should sometimes occupy positions 3 or 4 of figure 12 is not entirely clear. There may of course be a slight non-volatile acidosis in certain cases. A carbon dioxide acidosis, that is to say retention of carbon dioxide in the blood because of insufficient pulmonary ventilation, is theoretically possible and might be the explanation for those diagrams in which the *A* point falls in the 4 position.

That a carbon dioxide acidosis is a physiologic possibility has been shown by the work of Scott. This investigator determined the ventilation reaction of decerebrated cats to increasing amounts of carbon dioxide in the inspired air after the method used by Newburgh, Means and Porter. He found that when the inspired air contained 5 per cent of carbon dioxide an actual shift of the hydrogen ion concentration of the blood in the acid direction could be demonstrated. Scott found also that under these circumstances the bicarbonate reserve was increased but not in sufficient proportion to preserve the $\text{H}_2\text{CO}_3:\text{NaHCO}_3$ ratio at its usual value.

There are two possible explanations of why the ventilation should be insufficient in pneumonia: first, the obvious one that the pulmonary bellows is mechanically insufficient (this I will consider a little later); and second because of a decreased sensitivity of the respiratory center

towards its normal stimulus. Newburgh, Porter and I in 1916 showed that in experimental pneumonia there is a progressive decrease in sensitivity of the center as the disease advances. This was demonstrated by a progressive fall in the ventilation reaction to increasing amounts of carbon dioxide in the inspired air as the animals grew sicker. A series of such curves is shown in figure 22. Such a depression in the sensitivity of the center might be expected to give arterial points in the 4 position. To what it was due we are unable to say. It apparently was not the result of bacterial toxins, for animals

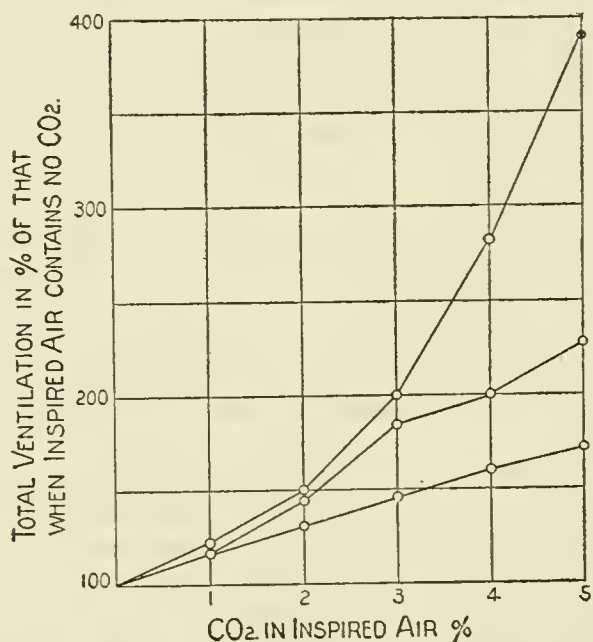


FIG. 22. VENTILATION REACTION TO CARBON DIOXIDE IN DOGS WITH PNEUMONIA

From Newburgh, Means and Porter. Upper curve, normal dogs. Middle curve, dogs moderately ill with pneumonia. Lowest curve, dogs very ill with pneumonia.

with septicemia caused by the same organism as that of the pneumonia animals failed to show any depression, nor could we produce it in animals with anoxemia due to carbon monoxide poisoning. Some later work by Porter and Newburgh suggests that it may in part at least be due to the effect of centripetal nervous impulses along vagus pathways. Personally I feel that anoxemia may have had something to do with it.

The work of Davies, Haldane and Priestley is interesting in this connection. They apparently demonstrated that resistance to breathing if excessive is capable of fatiguing the respiratory center. The progressive decrease in sensitivity of that center in pneumonia, discovered by Newburgh, Porter and myself, may also have been in part due to fatigue, particularly as our results showed that it was not the result of toxemia. The further work of Porter and Newburgh may then be interpreted as showing that impulses carried by the vagi are the stimuli producing the fatigue.

Of the blood flow in pneumonia, so far as I am aware, nothing is known. In the absence of cardiac insufficiency, however, it would seem reasonable to assume that it keeps pace with metabolism. A profound anoxemia may occur in pneumonia but it is of the anoxic,

TABLE 791

INTENSITY OF CYANOSIS	OXYGEN SATURATION OF ARTERIAL BLOOD	OXYGEN SATURATION OF VENOUS BLOOD
	<i>per cent</i>	<i>per cent</i>
0	95	77
+	89	68
++	83	59
+++	75	49
++++	48	17

not the stagnant type. It is undoubtedly due to incomplete aëration in the lungs, not to sluggish blood flow. The saturation of the venous blood may, it is true, be lower than normal but this is probably a reflection of the arterial unsaturation and not a result of stagnation.

Peabody in 1913 studying the venous blood of pneumonia patients found that in the terminal stages of the disease there was some reduction in saturation. Stadie in 1919 in patients with the broncho-pneumonia of influenza found abnormally low oxygen saturation in both arterial and venous blood. He also found that the intensity of the cyanosis varied in direct proportion to the degree of unsaturation of the arterial blood. The figures in table 3 are taken from the curves given in his paper.

As to why there should be anoxemia in pneumonia there are various possible explanations. Hoover in 1918 remarked upon the apparent

disassociation of dyspnoea and cyanosis in soldiers who had been gassed. In these men, who had both respiratory distress and cyanosis, the therapeutic administration of oxygen usually cleared the cyanosis but failed to relieve the dyspnoea. Hoover attributed this phenomenon to the presence of foam in the bronchial tree. Moisture in the air passages he believes usually exists in the form of foam. He points out that the presence of foam would interfere materially with both the intake of oxygen and the output of carbon dioxide. The foam would be made up of alveolar air bubbles. The rebreathing of expired air therefore would become very much greater than under normal conditions. This amounts essentially to a great increase in size of the functional dead space. Now when oxygen is given the foam bubbles gradually get saturated at the new tension and the rapidity of oxygen diffusion inwards is increased and the anoxemia and cyanosis relieved. The hindrance to carbon dioxide elimination, however, has not been removed, and hence the respiratory distress which Hoover apparently attributes in these cases to retention of carbon dioxide is unrelieved. This is a most fertile suggestion. It is quite conceivable that foam may play a part in the production of dyspnoea and cyanosis in patients with pneumonia. This would be especially true of the pneumonia of influenza in which there are often definite clinical signs of abundant moisture in the chest, and perhaps in certain other bronchopneumonias associated with extensive bronchitis.

Hoover goes on to discuss other types of cyanosis. In a frank lobar pneumonia with one or more lobes hepatized and the remainder practically normal, he points out that the cyanosis will not clear when oxygen is administered. This he believes due to the fact that the cyanosis is caused by the admixture of blood that has been through normal lung and been properly aërated with that which has been through hepatized lung and so not aërated, essentially the same idea as that of Haldane, Meakins and Priestley in the case of the uneven expansion of the lung in shallow breathing. Under such circumstances the administration of oxygen will accomplish nothing, for the blood which passed through normal lung was thoroughly saturated with oxygen when air was being breathed, and the rich oxygen mixture will no more reach the blood passing through hepatized lung than did

air. Hoover believes that exactly the same thing may happen when a portion of the lung is shut off from the outside air by a plug in its bronchus. In hydrothorax or pneumothorax, on the other hand, there usually is little or no cyanosis for the reason that the lung is collapsed and the flow of blood through it is greatly reduced, so most of the blood passes through the normal lung and hence there is no admixture of properly and improperly aërated blood. This last theory is borne out by the work of Balboni and myself on pneumothorax. We found that persons with a complete collapse of one lung increased their ventilation to increasing amounts of carbon dioxide in the inspired air in an entirely normal manner. Now if, in such persons, any great amount of blood were passing through a lung where it failed to get aërated, upon rebreathing hypercapnia would promptly be produced, and the curve of the ventilation reaction would be much steeper than normally, but such was not the case.

These several factors thus suggested by Hoover may individually or collectively play a part in the production of anoxemia in pneumonia and should be borne in mind when the various factors producing respiratory distress in any given case are under consideration. The admixture of unaërated blood from consolidated lung with aërated from sound lung tissue is probably unimportant. Gross has shown that the circulation through the solid portions of the lungs is, relatively to the sound portions, greatly diminished. It has also been shown by several observers that the anoxemia of pneumonia can be relieved by the administration of oxygen. This could not happen were it exclusively due to non-aëration in a consolidated lobe.

It is possible that shallow breathing is a factor in the production of anoxemia in pneumonia. This has been investigated by Meakins. The gas exchange and ventilation of four cases of pneumonia is given in his paper. The highest figure for the volume per respiration was 310 cc., the lowest 160 cc., and the average 255 cc. This is a distinctly low figure for the volume per respiration, practically half the normal, and is especially striking when contrasted with the total ventilation of these cases which varied between ten and thirteen liters, or in the neighborhood of double the normal resting ventilation. Two of Meakins' patients who died showed a progressive fall in the

volume of a single respiration until death, and two that recovered showed a prompt increase to about 500 cc. in the first day or two after the crisis.

The reason for the shallow breathing in pneumonia may be mechanical, chemical, nervous, or all three; chemical because of anoxemia produced perhaps in one of the two ways suggested by Hoover, and we have seen that in view of the experiments of Haldane, Meakins and Priestley, oxygen want produces shallow breathing and shallow breathing produces oxygen want; mechanical because of the fact that the vital capacity of the lungs must be reduced in pneumonia. I have made a few observations of the vital capacity in pneumonia and found a marked reduction. One man, for example, with a consolidation of the right upper and left lower lobes had a capacity of but 1.5 liters. He was a large man and should have had at least 4 liters when well. Nervous influences must play an important part in the production of shallow breathing in pneumonia, perhaps in part through fatiguing the center as I have already suggested, but more especially through the limitation of respiratory excursion to reduce pleural pain.

Whether the residual air is increased in pneumonia I believe is entirely unknown. I have never seen any data on the subject in the literature, and as a matter of fact, it probably would be impossible to get pneumonia patients to perform the respiratory gymnastics necessary to make such determinations. We do not know therefore whether in pneumonia there is any degree of pulmonary inflation or, as we might say, functional emphysema such as that occurring in decompensated heart disease.

Of the dead space also there seems to be a lack of information. I have encountered no papers giving the factors necessary to make the calculation. In Meakins' cases, however, the carbon dioxide content of the expired air was essentially normal and in those of other observers the alveolar carbon dioxide has been within normal limits or lower than normal. These facts taken together would be strong evidence against any increase in the size of the dead space, and furthermore as we have seen, the work of Haldane and of Henderson seems to show that, other things being equal, with shallow breathing the dead space is decreased. The pneumonia patient, then, unless he has anatomical

emphysema, probably is not in addition to his other respiratory troubles bothered with an enlarged dead space.

We may summarize then by saying that the pneumonia patient has increased ventilatory needs because of increased metabolism, anoxemia, and perhaps because of acidosis, and that at the sametime he has marked limitation of the efficiency of his bellows through restricted vital capacity and that for that reason he is forced to use a rapid shallow type of breathing which is not only uneconomical but may actually increase his anoxemia. His dyspnoea is therefore quite understandable. With the crisis the temperature falls and with it the metabolism, abdominal distention subsides, so too very likely does pleural pain, demands for ventilation decrease, supply increases, dyspnoea therefore rapidly disappears.

XIII. THE TREATMENT OF DYSPNOEA

In conclusion a few remarks on the treatment of the symptom we have been considering may not be out of place. Symptomatic treatment, when possible, should be based upon the known morbid physiology. The indications for the treatment of dyspnoea can be determined by the same methods that have been used to discover its causes.

First of all comes treatment directed toward reduction of the metabolic rate. This is seen most strikingly in treatment by rest, particularly in heart failure. Mere confinement to bed of course makes total metabolism closely approach basal and therefore greatly diminishes the work both of the heart and the respiratory organs, and consequently the dyspnoea. Morphine, by diminishing pain and restlessness, will act in a similar way. In the dyspnoea of hyperthyroidism, with or without organic heart disease, those measures which reduce the hyperthyroidism and therefore the metabolism by the same token diminish or abolish dyspnoea. The serum treatment of pneumonia, when successful in so far as it reduced fever and with it metabolism, would have a like effect upon dyspnoea.

Dyspnoea dependent on abnormalities in the ability of the blood to transport gases fall in the two main groups of those due to acidosis on the one hand and to deficient amount of available hemoglobin on the other. True non-volatile acidosis in my opinion constitutes an indication for sodium bicarbonate administration, unless the acidosis

is so mild that there is good prospect of speedy recovery without it. The acidosis of diabetes, for example, has been found to yield in certain instances to alkali plus insulin when it had not to insulin alone. The retention acidosis of nephritis also yields to alkali but of course only temporarily; even so it may be worth employing if the acidosis hyperpnoea is distressful.

The dyspnoea associated with low hemoglobin, which is of anoxemic origin, is relieved by whatever measures may restore the hemoglobin, as for example the transfusion of blood in severe anemia.

The dyspnoea due to slow blood flow, that is to say cardiac dyspnoea, is of course relieved by such measures as improve the muscular power of the heart, rest and digitalis. The effect of these agents is twofold, for not only is blood flow improved, thus diminishing anoxemia and the need for superventilation, but at the same time pulmonary congestion is decreased and consequently vital capacity is increased.

Of the agents which may be used to promote gas exchange in the lungs by far the most important is oxygen. This gas as a therapeutic agent is of great value. Its relatively infrequent use in clinics today is doubtless due to the lack of satisfactory modes of administration. Practically all anoxic anoxemia except that of congenital heart disease with right to left shunt and that due (if there is such) to the admixture of blood from solidified lung with that from normal lung will be either improved or abolished by oxygen therapy. Stagnant anoxemia of course is not affected by breathing rich oxygen mixtures, for the blood already is saturated in the lungs. The effect of oxygen therapy on cyanosis is a good criterion for distinguishing anoxic from stagnant anoxemia. The anoxemia of pneumonia Barach and Woodwell found could be nearly always materially relieved by oxygen therapy. So too could that of heart disease when of the anoxic type, that is to say when it was due to pulmonary causes, edema of the lungs, etc., and not simply to slow blood flow. The methods for giving oxygen continuously are being improved. The oxygen chamber at the Rockefeller Hospital marks an important advance in this direction. Simpler bedside apparatus has been devised (Barach) and undoubtedly in the near future an entirely satisfactory type will be evolved.

Of the use of drugs in the treatment of dyspnoea little more need be said. Morphine and digitalis have been mentioned. Morphine in large doses of course depresses the respiratory center and may lead to grave anoxemia from respiratory insufficiency. Used with care, however, there is no danger of this and in heart disease by providing complete rest or in pneumonia by improving the character of respiration through the relief of pleural pain it may be of great value. Of drugs which act as respiratory stimulants, that is to say which increase the excitability of the respiratory center or lower its threshold to its normal stimulus, caffeine is probably the most important. The chief therapeutic quandary in regard to respiratory stimulation is the determination of when it is truly indicated. Under what circumstances does one wish to stimulate the respiratory center? In morphine poisoning, of course, a direct indication for stimulation exists. Similar situations probably occur in disease but they are not easy to recognize. Theoretically the existence of a carbon dioxide acidosis together with anoxic anoxemia would be an indication. This condition may occur in pneumonia. Whether caffeine could materially relieve it or not I believe is not yet proved. Even if caffeine is indicated under such circumstances, oxygen is indicated as well, and it is possible that oxygen alone would be sufficient. The difficulty here in recognizing indications for treatment is because of one's ignorance of whether carbon dioxide acidosis, if it exists, is due to a depressed respiratory center or to a mechanical inability on the part of the pulmonary bellows to supply the ventilation required. The former might be helped by caffeine, the latter would not.

Adrenalin when used in bronchiolar spasm probably gives more dramatic relief to respiratory distress than any other agent. The indication is directly met by the agent employed. This can be shown by tracings of the breathing in which after the drug has been given the prolonged expiration is shortened, the volume per breath increased, and the vital capacity markedly increased in the space of a few minutes. The symptom, in other words, was due to an obstruction in the bronchiolar pathway. The drug abolishes the spasm and removes the symptom. Atropin works in a somewhat similar but less satisfactory way.

The procedures which improve the mechanical action of the thorax or which remove mechanical hindrances should never be forgotten. These include proper position in bed, that is an upright or semi-upright position when orthopnoea is present; comfortable and adequate support; also the relief of abdominal distention and of pleural pain.

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ALTERATIONS OF INTRAPLEURAL PRESSURE AND THEIR SIGNIFICANCE¹

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The danger of an open pneumothorax was apparently common knowledge to the ancient Greeks. Celsus (1) in discussing, the question of studying anatomy by dissection of living criminals, a practice in vogue in Alexandria in the third century before Christ, states:

It is indeed true that the abdomen, with which our argument is less concerned, can be opened while a man yet lives, but as soon as the knife reaches the thorax, and cuts the transverse septum, which is a membrane dividing the superior parts from the inferior and called diaphragma by the Greeks, the man at once gives up the ghost, and thus it is the breast and its viscera of a dead and not a living man which the murderous physician examines. He has thus but performed a cruel murder, and has not learned what the viscera of a living man are like.

Despite the antiquity of the knowledge that incisions cannot be made in the thorax with the same impunity as in the abdomen, our present knowledge of the processes involved are by no means complete, and in recent years even the idea that a thoracic opening is not so innocuous as an abdominal one has been repeatedly challenged. The problem, although a physiological one, has been given but scant attention by the physiologists. To the clinician it is a matter of prime importance to know in an exact manner what effects may arise from alterations in the intrapleural pressure; for some of the most common conditions are associated with some of these effects. To the thoracic surgeon it is essential that he be familiar at least with the grosser effects of a pneumothorax if he wishes to minimize his disasters.

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Under normal conditions each lung completely fills its corresponding pleural cavity except for the presence of a very small amount of fluid; there is, therefore, only a potential pleural space. But at the same time that each lung completely fills its pleural space its elastic tissue is tending to contract and to pull the lung away from the chest wall. It is therefore obvious that if the pressure between the lung and the chest wall be measured under ordinary conditions it will be found to be "negative," that is, it will be less than that of the atmosphere. The essential mechanical factor in inspiration is an enlargement of the thorax. This has a tendency to pull the chest wall away from the lung with the result that the intrapleural pressure becomes more "negative," that is, still less than that of the atmosphere. It is obvious therefore that the intrapleural pressure just at the end of a deep inspiration is lower, that is more "negative," than at any other time. Conversely, at the end of an expiration it is higher than at any other time, that is it is less "negative." Under ordinary normal conditions the intrapleural pressure does not become equal to or greater than the atmospheric pressure. Negative pressure at some time during the act of inspiration is necessary in order that air should rush down the trachea to fill the lungs.

Donders (2) was apparently the first to measure the intrapleural pressures in the human. At the end of quiet expiration he found a pressure of -7.5 mm. Hg. and at the end of inspiration -9 mm. Hg. Aron (3), in a living normal man, found at the height of quiet inspiration an average reading of -4.64 mm. Hg. and at the height of quiet expiration -3.02 mm. Hg. It is natural to expect that a normal variation in the intrapleural pressure will occur. There is therefore no absolute normal pressure reading for either inspiration or expiration. The literature on this subject is extensively reviewed in the exhaustive monograph of Emerson (4).

Abnormal conditions which may disturb the intrapleural pressures result from the presence of fluid, gas, or tumors in the pleural space. Although all of these will exercise pressure on the lung each operates in a slightly different manner. In the case of fluid the greatest pressure will be exerted at the bottom of the pleural space, in accordance with the laws of hydrostatic pressure. Gas, on the other hand, will exert an equal pressure in all parts of the pleural cavity, provided that

there are no adhesions to obliterate any of the space; and a tumor will exert its chief pressure wherever it happens to be located. It is therefore apparent that conclusions concerning the mechanics of the thorax which are based on observations made on the effects of fluid in the chest are not necessarily applicable to a consideration of the effects of air in the pleural space. The failure to recognize this fact has been responsible for much of the confusion in the literature on this subject.

To the surgeon the greatest interest is concerned with the effects of air in the pleural cavity because anyone who performs intrathoracic operations should be familiar with the effects which result from an opening in the chest wall. To the physiologist likewise, a study of the effects of a pneumothorax reveals important facts concerning both respiration and the circulation. Limitations of time will compel me to discuss chiefly in this paper the effects of a pneumothorax.

Until very recent times an accidental admission of air into the pleural cavity through a free opening was generally regarded with great dread because of the possibility of immediate death. Probably no other cause contributed to such a degree to retard the development of thoracic surgery as the fear of the consequences of making a free opening into a pleural cavity. To most writers on this subject any opening into a pleural cavity which permitted the free entrance of air signified an immediate collapse and loss of function of the lung on that side; the size of the opening was considered to be a matter of little or no consequence. Curiously confused with this idea was also the prevalent conception that, even in a normal chest, the mediastinal structures constitute a more or less rigid partition between the two pleural cavities. The literature on this subject is extensively reviewed in the articles by Emerson (5), Sauerbruch (6) and L. Mayer (7). Although the flapping of the mediastinal structures to and fro in a condition of open pneumothorax was frequently mentioned, there was but little realization of the extent to which the lung of the unopened pleura was handicapped.

A matter of great controversy at the present time, particularly among thoracic surgeons, is the question of just how much the opposite lung is affected when an opening is made in one pleural cavity. Much of this controversy unfortunately is concerned with opinions and conjectures rather than with actual pertinent data. Rational experi-

mental investigation of pneumothorax apparently began with Hewson (8) in 1767 who tried to produce it in animals by injury of the lung through the chest wall. Cruveilhier (9) found in 1836 that both pleural cavities of the dog may be opened simultaneously without a fatal result. This contradicted the earlier conception of Van Swieten that a bilateral open pneumothorax is necessarily fatal if the area of the combined openings is larger than that of the glottis. Since then until the last decade experimental studies of the effects of pneumothorax have been relatively few and sporadic.

In 1918, R. D. Bell and the writer (10), while members of the United States Army Empyema Commission, undertook an experimental investigation of the mechanics of pneumothorax, particularly with reference to the question of the effects of open drainage in cases of empyema. One of the methods of experimentation was the injection of air into one pleural cavity until a pressure of 10 cm. of water was obtained and observation of the resulting change of intrapleural pressure on the other side. The experiments were first conducted on human cadavers immediately after death and on normal dogs killed with ether. No marked difference between them was observed, when the thorax was normal in the sense that there were no adhesions and no thickening of the mediastinal structures. Accordingly experiments carried out on the living dog seemed applicable to living man. In the course of these experiments it was found that if an air pressure in one pleural cavity equal to that of 10 cm. of water is obtained, the pressure in the other cavity will be nearly the same, differing by only 2 or 3 cm. It was concluded, therefore, that from the standpoint of pressure relationships, the thorax can be considered practically as one cavity and that any alteration of pressure in one pleural cavity will show a change in the opposite one of practically the same degree. The sudden interruption of the work because of transfer to an overseas unit prevented further experiments with the use of higher pressures. It is only recently that a resumption of the work has shown that when higher pressures are used for experimentation there is a greater and greater difference between the pressures of the two sides in man and that therefore we were wrong in our former conclusion that any alteration of pressure in one pleural cavity will show a change in the opposite one of practically the same degree, although the previous

observations dealing with low pressures were correct. The details of these more recent experimental results will be discussed below.

Other conclusions which we drew from our earlier work were as follows:

1. Older conceptions are erroneous which imply that in pneumothorax only one lung is affected and that that lung is rendered functionless, regardless of the size of the opening.

2. On the contrary both lungs are affected; and, when a small pneumothorax is present, they are both affected to nearly the same extent. Moreover, a lung does not necessarily become functionless merely because there is a free communication between the pleural cavity of that side and the outside air. If such were the case then a bilateral open pneumothorax would promptly result in death from asphyxia. As a matter of fact, however, a bilateral open pneumothorax in an experimental animal is well tolerated and is compatible with life for an indefinite period, provided that the bilateral openings are not too large. I have found this fact to be true in both the dog and the rabbit.

3. A unilateral open pneumothorax will result in death if the opening be made sufficiently large.

4. The really important matter is the amount of air which enters through the opening at each inspiration and the ability of the individual to compensate for the encroachment on his respiratory surface which is caused by the pneumothorax. Roughly speaking this relationship becomes a question of the relation of the size of the opening and the value of the vital capacity of the individual. When the vital capacity is high a larger opening can be tolerated than when it is low; and no opening at all of any appreciable size can be withstood if the vital capacity is so low that it practically equals the tidal air. In other words, even a very small opening will result in a fatal asphyxia, if, before the opening is made, the vital capacity is so low that in spite of maximal inspiratory efforts only enough air is inhaled to satisfy the tidal air requirements. A mathematical expression was given to show how a calculation could be made of a maximum non-fatal opening in a given case if the vital capacity was known. This expression was based on the assumption that the pressures in the two pleural cavities in the normal chest were always nearly equal even in

the presence of a unilateral open pneumothorax (11). It was stated, however, that the value could be only an approximation because of the variability of some of the factors involved.

5. Obviously, the conclusions mentioned above are concerned only with relationships as they exist in a normal thorax, that is one without adhesions or inflammatory induration. If sufficient adhesions exist or if the mediastinal pleura has been made rigid by inflammatory induration, as for example in cases of chronic empyema, then the relationships discussed above no longer hold true, and no ill effects from pressure disturbances will result even if a very large opening is made.

Both the experimental observations briefly summarized above and the conclusions drawn from them have been questioned by several writers. There is, for example, a frequently quoted statement in the literature that the dog differs radically from the human in that normally there is a communication between the two pleural cavities of the dog, while in the human there is no such communication. This statement has been made by Matas (12), Duval (13), Yates (14) and others. More recently Snyder (15) has claimed that it is impossible to produce a unilateral pneumothorax in the dog because of the alleged communication between the two pleural cavities. I have examined 25 dogs with the particular purpose of finding out whether there are any anatomical openings in the mediastinal pleura and have never found any. It is interesting also that very recently Duval (16) and Matas (12) have retracted their former opinions about the presence of a normal communication between the two pleural cavities of the dog. Snyder (15) has stated that in the dog it is impossible to create a unilateral pneumothorax because the air passes at once from one pleural cavity through the mediastinum to the other one with the result that the pneumothorax is bilateral. He shows also that fluids pass readily from one side to the other, and he quotes Garland (17) and Calvert (18) as having previously called attention to this fact. Both Duval (16) and Matas (12), although now admitting the absence of any anatomical communication between the two pleural cavities in the dog, nevertheless maintain that in this animal the mediastinal septum is so thin and tenuous in comparison with that of the man that experimental observations of pleural pressures made in

the dog are not applicable to the human thorax. In our original article in 1918 we had concluded that they were applicable in all particulars. In reply to this phase of the controversy I wish to state that I have repeatedly produced a unilateral pneumothorax in the dog with marked deflection of the heart and mediastinal structures into the opposite pleural cavity and I have published the confirmatory x-ray reproductions (19). A reopening of the study, however, has convinced me that in many dogs the pneumothorax does soon become bilateral and that there is a considerable difference between even apparently normal dogs in the amount of resistance to pressure offered by the mediastinum. In some dogs simultaneous readings of the pressures in the two pleural cavities after the injection of air into one give results very similar to those in the human; but in others the results are such as would be expected in a bilateral pneumothorax. Even if one concludes that the dog may not be an animal entirely suited for the experimental study of *unilateral* pneumothorax, it would seem nevertheless that this animal would be suitable for the study of the phenomena associated with *bilateral* pneumothorax and that the results obtained in such a study would be applicable to a consideration of bilateral pneumothorax in the human. Even if one admits that the mediastinal septum in the dog is more easily stretched, more easily torn and more easily penetrated by gases and fluids than is the human, it is nevertheless perhaps not out of place to call attention to certain irregularities in the methods of experimentation. If one wishes to measure the pressure in the opposite pleural cavity after injecting air into one, it is necessary to permit the entrance of a little air into the opposite cavity in order to have a cushion of air around the cannula which will permit a reading. If this is not done, the lung will usually force itself against the end of the cannula and prevent any oscillation of air within the cannula and tube. One starts therefore with some degree of bilateral pneumothorax which probably results in the retraction of the lung away from the mediastinal pleura. The final result is that if the lung is retracted away sufficiently the mediastinal septum will be hung in space at its edges without the buttressing support of the lung against it. It is obvious then that a thin mediastinal partition will be ruptured more easily under such circumstances than under the conditions which are

presented in the case, for example, of a surgical unilateral pneumothorax. For in the latter the only air which can be present in the opposite pleural cavity will be that which may have diffused through the mediastinum; moreover the mediastinum will be everywhere supported by being buttressed against the opposite lung. What we are really desirous of knowing, therefore, is not merely what the resistance of the mediastinal partition is but also how much is the other lung compressed by a force transmitted through the mediastinum. Nearly all of the experimental observations that have been published are really concerned only with the question of the resistance of the mediastinum.

Even the admission on my part that experimental observations on unilateral pneumothorax in the dog may not be applicable to man in their entirety does not in any way annul the chief conclusions to be drawn from our earlier work, although these have been severely criticized by Duval (13). He states, for example, that the size of the pleural opening is not a matter of great importance as regards the possibility of causing death. He is a strong advocate of a very large incision for the expressed purpose of making a total unilateral collapse of the lung to permit no respiration at all on that side and no "ventilation" of the pleura. He desires to make that lung completely airless. He states that this procedure also prevents the circulation of "pendelluft," a term used by Brauer to indicate the theoretical transfer of air from the unaffected lung into the partly compressed lung at expiration and the rebreathing of this air into the uncompressed lung on inspiration. Brauer's idea was of course that this phenomenon was an important factor in the production of asphyxia because this air would have its oxygen diminished besides containing an excess of CO_2 and would also prevent the inspiration of that much fresh air. Duval claims that the thorax of the bovine class of animals resembles that of the human and that an animal of this class should be used for experiments instead of the dog. He states that an experiment which he performed on a calf lends support to his ideas. He considers that in order to make a very large opening safe all that is necessary to do is to immobilize the corresponding side of the chest and diaphragm by means of a powerful automatic retractor and to express from the lung its residual air. To accomplish this he delivers the lung out of the

wound. He also recommends enveloping and progressively compressing the lung with warm gauze compresses. He regards the "ventilation" of the pleura as the most serious and harmful factor resulting from an open pneumothorax. By this he means presumably the passage of air in and out of the pleural cavity. In his criticism of our work he makes no reference at all to the importance of the vital capacity of the patient, a point which we strongly emphasized. I (20) have already replied to Duval's criticisms in a separate article; but I shall discuss some of his points and conclusions here.

It is rather generally admitted that a unilateral open pneumothorax may sometimes have disastrous results. Duval's conclusions differ diametrically from mine as to the reason for these disasters. He considers them as being due to the fact that the opening is not large enough, whereas on the contrary, the evidence seems to me to point to the fact that they arise because in a given case the opening has been too large. From the practical standpoint of surgical methods it is of the utmost importance to settle this question definitely. For that reason it will be well to analyze with some care the effects of a pneumothorax in order to gain a clearer insight into this important question.

The majority, if not all, of investigators who have used animals for the experimental study of pneumothorax have observed a change in the character of the respirations after the creation of an open pneumothorax. Likewise most surgeons of wide experience in thoracic work whose observations have been carefully made have noticed a change in the character of the respirations in the human in the presence of an open pneumothorax. Usually the first alteration is an increase in the depth of inspiration (an increase in amplitude), followed later by an increase in the rate. In an earlier paragraph I have already pointed out that the act of getting air into the lungs consists chiefly of enlarging the thorax, a procedure which necessarily reduces the intrapleural pressure (makes it more negative and thus causes air to rush down the trachea to inflate the lungs). We should expect similarly that whenever a sudden interference with the intrapleural pressure occurs which would make it equal to that of the atmosphere, the natural compensatory mechanism to offset this would be an attempt to create again a negative intrapleural pressure

in order to get air into the lungs. The simplest way to accomplish this is to enlarge the thorax still more in an effort to reduce its interior pressure. This is just what happens then as an automatic effort to counteract the sudden increase in the pleural pressure. This reveals itself in an increase in the depth or amplitude of the respiration. For purposes of simplicity we can compare in this respect the lungs and pleural cavities with any object containing a gas, as for example, a toy balloon. If the balloon should suddenly be doubled in size, or if the gas were suddenly transferred to a balloon of twice the size of the original one, the pressure of the contained gas would be only one-half of what it had been originally. In a structure like the thorax which is constantly changing its size, an opening communicating with the outside air does not necessarily imply that the pressure within is always equal to that of the atmosphere, especially if the opening is a small one. This has been a common source of confusion. During inspiration the intrapleural pressure is usually negative and during expiration it is positive, if the opening is not too large.

Haldane and Poulton (21) have shown that if an individual is placed in an atmosphere containing 4 to 5 per cent of CO_2 his respiration increases in depth and little by little increases in rapidity. On the other hand, if the amount of inspired oxygen is suddenly reduced below 14 per cent there is also at first an increase of amplitude of respiratory movements which they think is to be attributed to the fact that the anoxemia, by lowering the threshold of the respiratory center for CO_2 provokes the expulsion of CO_2 from the body. At the same time the rate is accelerated as the result of the anoxemia. Finally the respiration becomes periodic. If one continues to reduce the percentage of oxygen in the inspired air to 10 per cent the respiration becomes superficial and finally death occurs. Haldane, Meakins and Priestley (22) more recently have shown that if, instead of lowering the partial pressure of oxygen in the inspired air, one leaves the air at its normal composition but diminishes the amount of air entering the lungs sufficiently to produce a superficial respiration, the respiration soon becomes periodic. Periodic respiration is known always to be due to lack of oxygen. The anoxemia therefore induces superficial respiration which itself maintains the state of anoxemia. A serious vicious circle is therefore established.

Now Duval states that in a unilateral open pneumothorax dyspnea exists as long as the corresponding lung respire partially and "dances" a little in the thorax, that is, as long as the external air strongly ventilates the pleura, but that as soon as that lung is put into condition of complete collapse the dyspnea disappears. I have not been able to confirm this observation although, in accordance with Duval's recommendation, I used a calf for experiment. Moreover Dautrebande and Spehl (23) fail to agree with Duval in spite of the fact that they used rabbits, animals which like the bovine group, Duval recommends for experimental use because of their similarity to the human. In fact, in direct contradiction to Duval's claims I found that a calf dies if a large unilateral opening is made in the chest. It is difficult to understand how there could be any disagreement on a point so easy to demonstrate experimentally. When a large opening is made, however, the respirations soon become much more superficial, a fact which perhaps Duval has interpreted as a reduction in the dyspnea. But this is not to be regarded as a favorable sign. On the contrary, as Dautrebande and Spehl have shown in their excellent article, it should be looked upon as a danger signal because it indicates an anoxemia. This conclusion would agree very well with the work of Haldane and his pupils to which reference is made above. Accordingly Dautrebande and Spehl state that they regard Duval's conclusions as erroneous.

As has been mentioned above, Duval regards a large opening in the chest as less dangerous than a small one. My own opinion, however, based on many experimental and clinical observations is that, other things being equal, the larger the opening the greater the danger to life. The reasons for this conclusion are many. In the first place, it has been a frequent observation that animals will endure a bilateral open pneumothorax almost indefinitely if the openings in each side of the chest are not too large. In the case of dogs and cats of ordinary size, for example, if a small trocar is inserted into each pleural cavity the animal will go on for hours with often no evidence of any disturbance except for an increase in the amplitude of the respiratory movements, and this disturbance will promptly disappear after the withdrawal of the trocars. If, however, too large openings are made the animal will soon die with the picture of asphyxia. It seems to

me that this simple experiment permits several important conclusions. It shows first of all that a lung does not become functionless merely because outside air is allowed free entrance to the pleural cavity. It shows also very clearly that the important consideration is the size of the openings. Other subsidiary factors in the production of death, to which great importance has been attached by some, such as "pendelluft" and the fluttering of the mediastinum, are ruled out in a case of bilateral open pneumothorax, induced by openings of the same size, which has proved fatal; for in such a case, since the pressures are equal in both pleural cavities at all times, there will be no to and fro motion of the mediastinum and also no chance for the occurrence of "pendelluft," and yet the animal dies. Now when we come to consider the question of a unilateral open pneumothorax we find that practically the same phenomena occur. The principles involved are substantially the same as are involved in a bilateral opening because the effects of a unilateral opening are not confined to only one lung but concern both lungs. The details, however, are slightly different, because, as has been said above, the pressures in the two pleural cavities during a unilateral open pneumothorax are not quite the same and because there is some to and fro movement of the mediastinum, etc. Nevertheless, in view of what we have just said about a bilateral open pneumothorax, it seems reasonable to suppose that with a unilateral opening such questions as the fluttering of the mediastinum and the action of "pendelluft" are of less importance than the direct effects on the lungs caused by the altered pressures. This brings us back again to the importance of the size of the opening.²

² In a recent article Lundsgaard and Van Slyke (24) invoke an idea similar to that of Brauer's "pendelluft" as an explanation of cyanosis in unilateral open pneumothorax. They quote experiments by Sackur (26) who, after producing a supposed collapse of one lung in rabbits and in a dog by unilateral open pneumothorax, found a diminished content of oxygen in the arterial blood. On the basis of these experiments they conclude that cyanosis in acute unilateral open pneumothorax is produced "in the same manner as when the air passages to one lung are blocked, viz., by blood flow through unaerated lung space." In considering the pressure effects of open pneumothorax as if only one lung were concerned, they show that they are unaware of the fact that these effects involve both lungs. This leads then to the assumption that the frequent absence of cyanosis in spontaneous pneumothorax in tuberculosis is due perhaps to extensive thrombosis of the vessels in the lung on the involved side, a condition which they infer would prevent cyanosis because a relatively small amount of blood would pass through that lung. It seems to me just as

The usual attempt to compensate for the increased pleural pressure is to take a deeper inspiration, as has been discussed above. Theoretically all that is necessary to do to prevent a fatal asphyxia is to get enough air into the lungs to satisfy the tidal air requirements. This can be accomplished of course in spite of an open pneumothorax provided that too much air does not enter the pleural cavity with each inspiration. If so much air enters the pleural cavity through the open pneumothorax wound that pulmonary inflation is sufficiently compromised to prevent the possibility of inhaling enough air into the lungs to maintain life, then of course a fatal asphyxia will occur. Fortunately here, as in other respects, nature has made provision for a generous reserve. The tidal air requirements ordinarily constitute only about one-eighth to one-twelfth of the total amount of air which can be inhaled if a maximal effort is made. This latter amount of air represents what is known as the vital capacity. It is apparent therefore that a very considerable amount of lung tissue can be made functionless and yet permit the individual to take in enough air to maintain life by increasing his respiratory movements. This then would seem to be the explanation of the well-established fact that life is compatible, in a vigorous adult, with a relatively large unilateral opening and even with bilateral openings whose combined areas greatly exceed the area of the opened glottis. A common error in the older literature on pneumothorax was that a lung becomes collapsed and functionless if a pleural opening as great or greater than the diameter of the glottis is made. A person who cannot make so great a respiratory effort cannot resist so large a pleural opening as the one who can make a great inspiration; likewise it becomes apparent that the same individual, if he has any serious impediment to his respiration, will die from a pleural opening which would not be fatal if the impediment were absent. In other words, therefore, the vital capacity of the individual is of great importance in determining the size

logical to assume that the explanation of the absence of cyanosis in such a case lies in the fact that because of the presence of adhesions and fixation of the mediastinum there are no serious pressure effects on the opposite lung and that therefore actually a less amount of lung tissue is involved than if no adhesions were present, to say nothing of the altered relationships of the heart and large vessels when a pneumothorax is induced in the absence of adhesions.

of a pleural opening which is compatible with life. One who has a large vital capacity can withstand a larger pleural opening than one whose vital capacity is low. By considering what conditions lower the vital capacity we can understand readily that a large pleural opening is likely to be fatal in an individual whose respiratory muscles are weak, who has an uncompensated heart, who has an extensive pneumonia, a pulmonary edema, an acidosis, a deep general anesthesia, etc. Indeed, if he is so dyspneic that he just succeeds in maintaining life in spite of maximal respiratory effort, in other words if his vital capacity practically equals his tidal air requirements, he may succumb to the effects of even a very small pleural opening.

In some previous publications I (26, 27) have called attention to the fact that on purely theoretical grounds, even if an alteration of pressure in one pleural cavity induced a change in the other of the same degree it can be calculated that a pleural opening can be withstood of such a size that it would have seemed impossible before the war experience. For example, on the basis of our calculation, a man with a high vital capacity (7180 cc.) could support life with a unilateral opening as large as 15.6 square inches, or 101 square centimeters. These calculations, however, are not strictly accurate but are only approximations because of the variability of several of the factors. As a matter of fact, since the pressures in the two pleural cavities are not equal when there is a great increase in one, it is seen that in some individuals with high vital capacities a unilateral opening considerably larger than 101 square centimeters may be compatible with life. It really is of little importance to know exactly in any given case how large an opening can be made. The important thing to realize is that those whose vital capacities are low, especially those who are already dyspneic, may die as a result of even a small opening unless measures are taken to prevent serious consequences.

The most important aid in minimizing the harmful effects of an open pneumothorax is to close the opening. Closure of the opening leads to an almost instantaneous improvement in the respiration. The explanation of the relative innocuousness of a closed as compared with an open pneumothorax is apparently as follows: When an opening is closed, particularly if it is closed at the end of expiration, there is not enough air remaining in the pleural cavity to encroach on the breathing

surface of the lungs sufficiently to prevent the establishment of a negative intrapleural pressure by a slight increase in the depth of inspiration. There is no longer an active competition between the passage of air down the trachea on the one hand and the entrance to air into the pleural cavity on the other hand. The impossibility of getting the requisite amount of tidal air into the lungs does therefore not occur. Experimentally, however, a closed pneumothorax can be made to cause a fatal asphyxia if enough air is injected into the pleural cavity. Here again the process seems to consist fundamentally of increasing the intrapleural pressure to such an extent that it becomes impossible to create a negative pressure in spite of maximum respiratory efforts; the necessary amount of tidal air cannot be obtained, with the inevitable result that asphyxia occurs. Again, moreover, if the vital capacity is low, it is easier to produce death with a closed pneumothorax for reasons which have been made sufficiently clear in the discussion given above. Doubtless some of the catastrophes which have occurred in the production of artificial pneumothorax for therapeutic purposes are explainable on the fact that patients with low vital capacities and without adhesions have been given too much air. An amount of air which might be well tolerated by a patient with a high vital capacity might be fatal in one whose vital capacity is low. The prompt withdrawal of air in an alarming case will usually improve the dyspnea and prevent death. It is an interesting fact that Duval and others who have criticized our theory of the action of pneumothorax recommend strongly the use of measures which tend to reduce the size of the opening when performing operations on the chest. They therefore unconsciously support our theory while at the same time they attack it. For example, they frequently deliver the lung out of the incision; they also pack in gauze compresses. They lose sight of the fact that the size of the incision and the size of the opening are not synonymous when anything is in the incision, no matter whether it is a delivered lung, a gauze compress, the surgeon's hand, instruments or what not. A hole that is either partly or completely plugged obviously cannot be as large as if it contained nothing. Other aids in combating the asphyxial effects of an open pneumothorax are the administration of either oxygen or carbon dioxide, as recommended by Dautrebande and Spehl. to break up the vicious circle

which occurs when the anoxemia appears. Oxygen will supply directly the thing that is most needed, but the administration of carbon dioxide will accomplish the same purpose indirectly by stimulating deeper inspirations and thereby the taking in of more air.

Since the explanation of the action of pneumothorax which I have summarized here has not met with universal acceptance, it will be worth while to see how well it harmonizes with the clinical facts which were brought out particularly during the war. One fact which perhaps stood out above all others in relation to war wounds of the chest was the unanimous agreement that it was important to close sucking (open) wounds promptly. The high mortality attending open wounds was attributed by some to the increased risk of infection. This was undeniably an important factor, but yet, according to Duval (28), 50 per cent of all fatal lung wounds died within the first day. It seems improbable that a severe enough infection could have manifested itself so soon as to be responsible for so high a mortality and likewise it is doubtful if hemorrhage could have caused most of the deaths. Is it not more probable that many, if not most of these, deaths occurred as a result of the pressure disturbances incidental to an open pneumothorax? This point is discussed at more length in our original article published in 1918. Again the fact that living men were often seen with wounds of the chest wall which were so large that they would have seemed before the war to be incompatible with life does not seem strange in the light of the theoretical discussion presented above. Finally also, in the case of acute empyema, it would seem to me that nothing more clearly substantiates the truth of the principles here set forth than the remarkable reduction in the mortality which followed the abandonment of the practice of early open drainage in the acute pneumonic stage of the disease. The explanation of the remarkable decrease in mortality accomplished by the postponement of open drainage seems easy in the light of the discussion given above. During the stage when pneumonia is present the vital capacity is low (11). In some cases it was actually so low that in spite of maximal respiratory efforts it was difficult for the patient to take in enough air to maintain life. Moreover, at this time there was usually no walling off of pus in the pleural cavity. An open drainage, therefore, at this stage of the disease carried with it all the dangers of an open pneu-

mothorax at a time when, in many cases, the vital capacity was so low that an opening of any size would have been fatal. Later, however, after the pneumonia had cleared up, not only had the vital capacity increased but also usually the pus had become more or less circumscribed by adhesions so that a drainage opening did not create a free pneumothorax. These remarks hold true especially for the type of pneumonia and empyema due to the hemolytic streptococcus. I believe the principle of avoiding an open drainage during the stage of acute pneumonia is now universally accepted in this country. At Camp Lee we saw the mortality drop from more than 40 per cent to less than 5 per cent by carrying out this principle. Stone (28), at Fort Riley, showed a correspondingly marked reduction in the mortality after the early open drainage during the pneumonic stage was abandoned. The tabulation of his results is as follows:

1. *First series.* Early operation (October 20, 1917, to January 21, 1918). 85 cases; mortality, 61.2 per cent.

2. *Second series.* Early aspirations and late operation (January 12 to August 10, 1918). 96 cases; mortality, 15.6 per cent.

3. *Third series.* Early aspirations and later operation (October 18, 1918, to February 14, 1919). 94 cases; mortality, 9.5 per cent.

At the St. Louis Children's Hospital, since September, 1919, we have treated 90 cases of acute empyema by a plan of repeated aspirations during the pneumonic stage to be followed by free drainage when necessary. In this series there have been 11 deaths, but in 10 of these there were serious complications, such as suppurative mastoiditis or meningitis, etc. This mortality of 12 per cent compares very favorably with that of 54 per cent given by Holt (29) in a series of 126 cases in children reported before the war. The empyema records of the St. Louis Children's Hospital before the war are not available.

There are also now a considerable number of investigators who have carried out experiments since the publication of our paper in 1918 whose results in the main agree with ours. For example, Stivelman, Hennell and Golembe (30) state that "Graham and Bell touched the heart of the subject. . . . In the presence of a flexible mediastinum, the intrathoracic equilibrium in pneumothorax is very delicately adjusted, and any disturbance in the intrathoracic pressure on the treated side will have a proportionate effect on the intrathoracic pres-

sure on the untreated side." Lenhart (31), in experiments on rabbits, found that unilateral open pneumothorax causes an intense dyspnea, provided the anesthesia is not too deep, and death, if unrelieved by closure of the thoracic opening. The volume of air respired per minute is decreased despite the increased respiratory effort; the excretion of carbon dioxide is impaired, often to a great degree; the respiratory quotient is constantly reduced; there is an increase in the total carbon dioxide content of the blood, and at the same time an increase in the hydrogen ion concentration. Finally he states that his opinion concerning the alteration of pleural pressures on both sides as a result of a unilateral open pneumothorax is in agreement with ours. Simon (32) has also come to practically the same conclusion as a result of his experiments.

In the discussion here of the effects of pneumothorax I have dealt particularly with the effects on respiration. But there are also other effects, for example, on the circulation. Sauerbruch summarized these as follows: "In pneumothorax the aspiration of the heart fails; a stasis results in the venous system. Measurements of the venous pressure in the femoral vein give in fact an increase of the pressure." As a result of asphyxia there is of course a rise in the arterial blood pressure; this is a general result of asphyxia and has no particular relationship to the question of asphyxia produced by pneumothorax. Yates has called attention to the importance of the pulmonary circulation in the inflation of the lung. He states that

Wherever intrapulmonary pressure exceeds intravascular tension a proportion of the blood destined under normal conditions to reach the compressed lung area is immediately diverted to adjacent channels offering less resistance. The capillary network that surrounds the air cells spreads, as its component vascular tubules are thus elongated by overfilling. If air passages are free and lung tissue is elastic, the result is increased inflation in the portion of lung to which extra blood has been delivered, a compensatory physiologic emphysema. On the contrary a temporary stoppage of pulmonary circulation produces atelectasis; permanent stoppage causes atrophy in the lung affected. In short, the pulmonary circulation is the most important factor influencing pulmonary inflation, and fluctuation in pulmonary inflations is accompanied by variations in the resistance to blood flow in the pulmonary circulation.

These are important considerations. They are, however, secondary results of the alteration of pressure produced by the pneumothorax. Moreover, so far as the atelectasis resulting from a temporary stoppage of circulation is concerned, time is necessary for its accomplishment. The instantaneous effects on respiration which follow the creation of an open pneumothorax and the instantaneous improvement

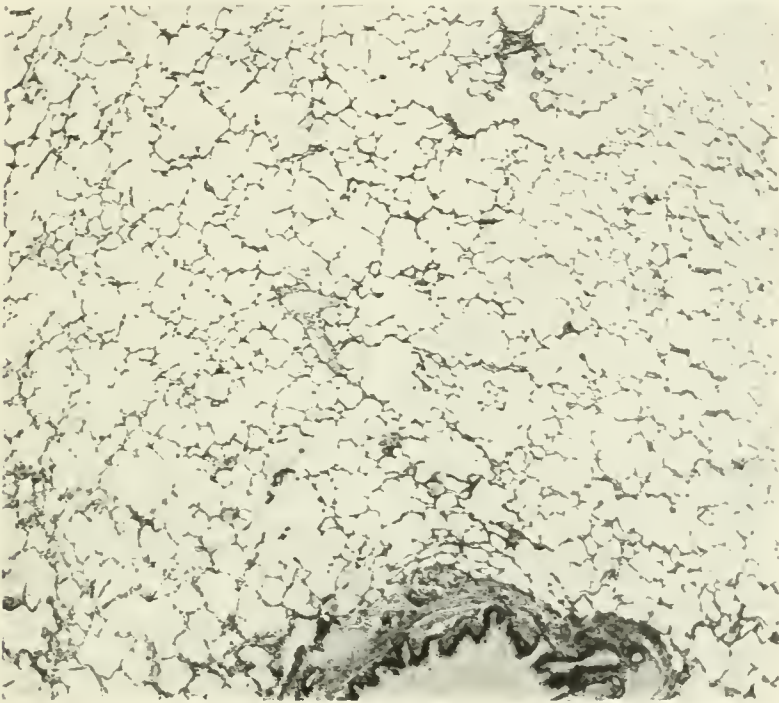


FIG. 1. SECTION OF NORMAL PORTION OF RABBIT'S LUNG. FORMALIN FIXATION

in the 'dyspnea which follows the closure of the opening must be explainable on the basis of the immediate pressure effects exerted on the lung parenchyma rather than on the more slowly developing effects of disturbances of the lung circulation. The ideas of Yates, however, are of great interest in connection with the mechanism of the production of compensatory emphysema. He himself tries to explain Skodaic resonance on the basis of an increased blood supply and therefore, as he reasons, an increase in the size of the alveoli, in the part of the

lung which shows the hyperresonance. When this idea is put to the test of experiment, however, it is found that distention of the capillaries induces a diminution rather than an increase in the size of the alveoli. Experiments were carried out by H. J. Davis and myself

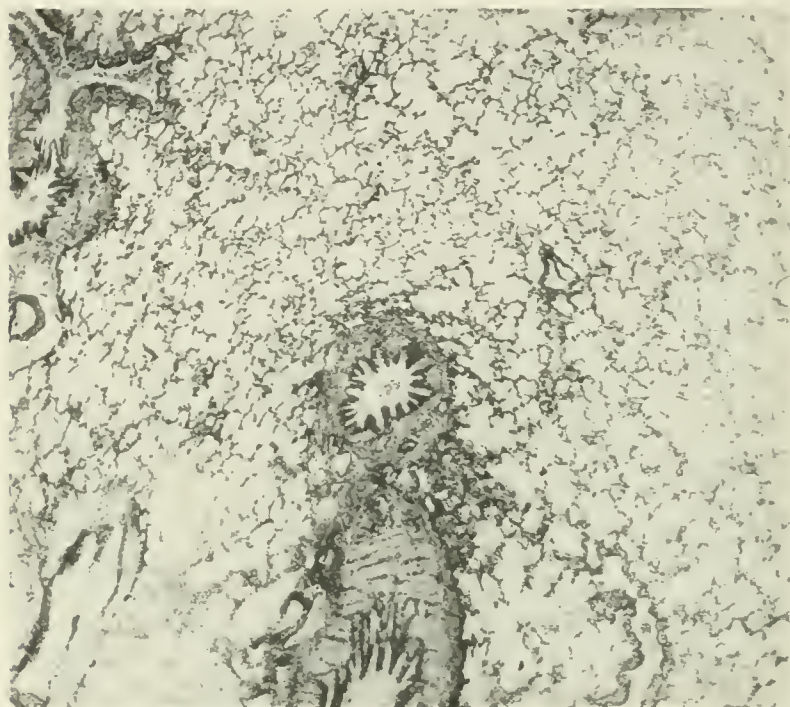


FIG. 2. SECTION OF ANOTHER LOBE OF SAME LUNG AFTER INJECTION OF THE CORRESPONDING BRANCH OF THE PULMONARY ARTERY WITH PHYSIOLOGICAL SALINE SOLUTION

Immediately after the injection the lobe was fixed in 10 per cent formalin. The alveoli are contracted instead of being distended, as compared with the normal control in figure 1.

(33) on a series of dog's lung. Immediately after death from ether the chest was opened and the lower lobe of each lung was removed. One of the principal blood vessels to the lobe was then injected with physiological sodium chloride solution, in some cases an artery and in other cases a vein. A tight ligature was then placed around the hilus of

the lobe, including both the vessels and the bronchus, and the lobe was quickly immersed in 10 per cent formalin. In every instance the alveoli of the lobe whose capillaries were distended were much smaller than those of the other lung. This was found true not only for normal lungs but also for those with pneumonia. It seems probable therefore that Skoda's resonance in pneumonia cannot be explained on the basis of an increased blood supply to the part. The accompanying illustra-

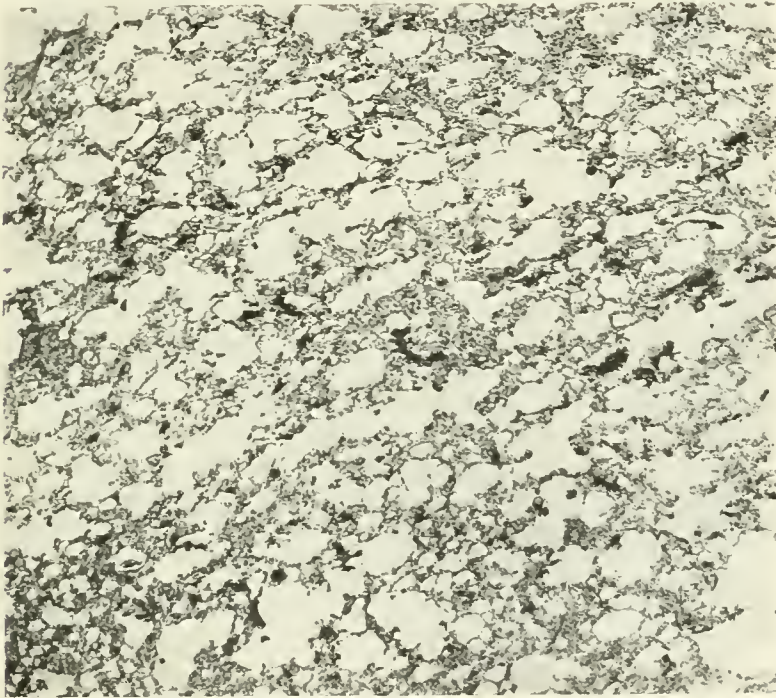


FIG. 3. RABBIT'S LUNG WITH BRONCHOPNEUMONIA. FORMALIN FIXATION

tions (figs. 1 to 4) show that the size of the alveoli is diminished as a result of the distention of the capillaries. Conversely it might be expected that when the alveoli are greatly distended the capillaries are diminished in size. That this result actually happens has been shown by the interesting work of Chillingworth and Hopkins (34) who studied the question by placing dogs in a specially devised body plethysmograph which included the entire dog with a tracheal cannula project-

ing outside. These investigators found that when the lungs are artificially distended by diminishing the plethysmographic pressure various results may occur as follows: (1) With slight distention there is a slight rise in the carotid blood pressure due to the freer passage of blood to the left heart. (2) With moderate distention a fall in caro-

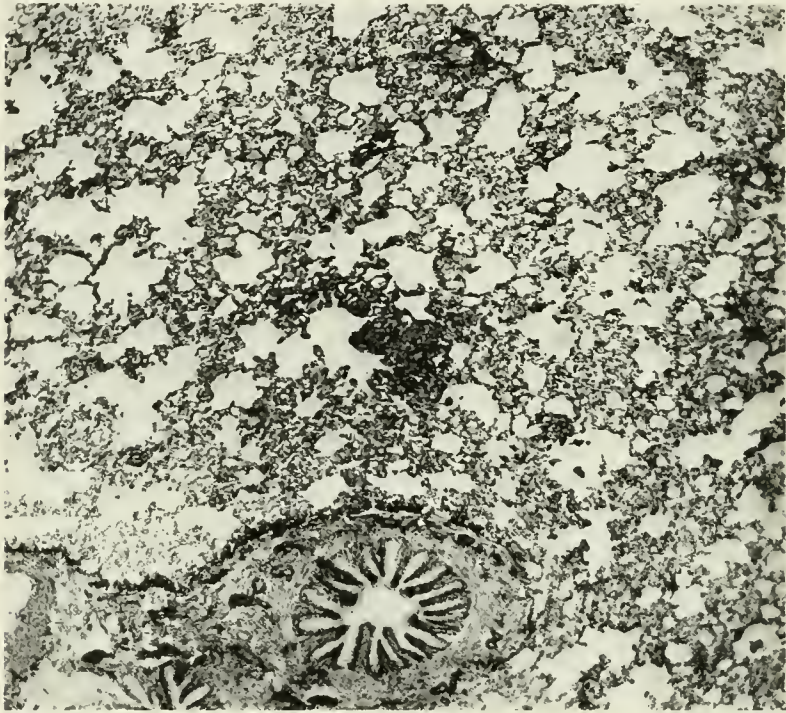


FIG. 4. ANOTHER LOBE OF SAME LUNG AFTER INJECTION OF THE CORRESPONDING BRANCH OF THE PULMONARY ARTERY WITH PHYSIOLOGICAL SALINE SOLUTION

The method used was the same as described under Fig. 2. The alveoli also of a pneumonic lung are reduced in size when the capillaries are filled.

tid pressure occurs. (3) With full distention the carotid pressure falls approximately to zero. (4) With overdistention, causing emphysema and pneumothorax, there is less of a fall of carotid pressure than that produced by less distention. (5) The fall in the carotid blood pressure caused by moderately diminished plethysmographic pressure is due to failure in the supply of blood to the left heart, a failure

which is due to pressure exerted almost directly upon the small pulmonary vessels.

Other effects of an open pneumothorax are marked loss of heat and the great danger of infection. Sauerbruch found that in unanesthetized rabbits in which he had established an open pneumothorax at a room temperature of from 20° to $22^{\circ}\text{C}.$, the body temperature sometimes dropped as much as $3.5^{\circ}\text{C}.$ within forty-five minutes. In dogs, also unanesthetized, within a half-hour he observed the body temperature fall $2^{\circ}\text{C}.$ after making a pleural opening and rise again $1.6^{\circ}\text{C}.$ within an hour after closing the opening. He also made the very striking observation that the heat loss in open pneumothorax is greater than in an extensive laparotomy with eventration of the intestine for the same length of time. For example, a dog's temperature fell only $1^{\circ}\text{C}.$ after having his intestine lying out of his abdomen for forty-five minutes, and a rabbit subjected to the same experiment showed a heat loss of only $1.3^{\circ}\text{C}.$ after forty-five minutes.

The danger of a general infection of the pleura in open pneumothorax is very great and is much greater than is the danger of a general infection of the peritoneum in the case of an open wound of the abdomen. There are probably two reasons for this fact; one is that with the suction of air through the pleural opening large numbers of bacteria are drawn in, and the other is that, with the lung retracted away from the chest wall, there is not the same possibility of sealing the opening as is the case in the abdomen by the action of the omentum and the viscera.

With reference to this discussion on the effects of pneumothorax the question naturally arises, Is positive pressure necessary in the performance of intrathoracic surgical operations? In cases without adhesions or without induration of the mediastinum it will be possible to make a large pleural incision in an individual whose vital capacity is high, particularly if those measures are applied which have already been discussed, namely, the delivery of the lung out of the incision, the insertion of gauze packs into the wound, etc., in other words, the use of any plan which minimizes the size of the actual opening. On the other hand, in patients with low vital capacities and without adhesions, the use of a large pleural incision invites disaster unless the means are at hand to institute some degree of positive pressure. In any case, it



FIG. 5. SPONTANEOUS BILATERAL PNEUMOTHORAX IN A DOG KILLED WITH ETHER
Picture made twelve hours after death



FIG. 6. SPONTANEOUS BILATERAL PNEUMOTHORAX IN ANOTHER DOG TWO HOURS AFTER DEATH

seems to me, an intrathoracic operation should not be undertaken unless one is prepared to administer positive pressure. No cumbersome apparatus is necessary for this. An ordinary nitrous-oxide machine with a tightly fitting face piece, as suggested in 1910 by Bunnell (35), is all that is necessary. Such a device has the additional advantage that not only can positive pressure be applied but that also oxygen can readily be supplied quickly in case of the development of a serious anoxemia. In any case, relative speed in operating is highly desirable. Not only does a quick operation diminish the chance of a serious anoxemia but it also minimizes the loss of heat through the incision, a point which is very important in thoracic work.

It is now well known that the air in a closed pneumothorax disappears completely within a few weeks and sometimes within a few days. The early pioneers in the therapeutic use of artificial pneumothorax, as for example Forlanini, thought it would be advisable to use nitrogen because supposedly it would disappear less rapidly. Later work, however, showed that, owing probably to the diffusion of gas from the lung into the pleural cavity and vice versa, the gas in a pneumothorax, even if pure nitrogen originally, soon came to have the composition of alveolar air. For that reason it is now customary to use air for artificial pneumothorax rather than nitrogen because of its greater convenience. This transfer of gases back and forth between the lung and the pleura is analogous to a process of respiration outside of the lung. It resembles therefore somewhat the respiration in the alimentary canal which was studied by Woodyatt and myself (36) in 1912. Despite the interest attached to the phenomenon of the transfer of gases from the lung into the pleural cavity and its reverse, it has never received the attention which it deserves. The diffusion of gas into the pleural cavity is so rapid after death that, at least in the case of the dog, an easily demonstrable bilateral pneumothorax may be seen in x-ray plates taken one hour after death. In one instance, from one pleural cavity of a 10 kilo dog 32 cc. of gas were obtained twenty-four hours after death, an analysis of which by Dr. P. A. Shaffer showed 17.5 per cent of CO_2 (figs. 5 and 6).

Another interesting problem dealing with alterations of the intrapleural pressure concerns the question of the formation of pleural exudates. The rapidity with which fluid accumulates in cases of

infection of the pleura with the hemolytic streptococcus and the fact that dyspnea is often a prominent feature of streptococcal pneumonia suggested the idea that fluid might actually be sucked out of a very edematous pleura by the negative pressure during the forced inspirations. In this type of pneumonia, as is well known, the dyspnea is often so marked that maximal inspiratory efforts are required in order to take in enough air to maintain life, due largely to an actual obstruction of the air passages with the products of inflammation. The con-

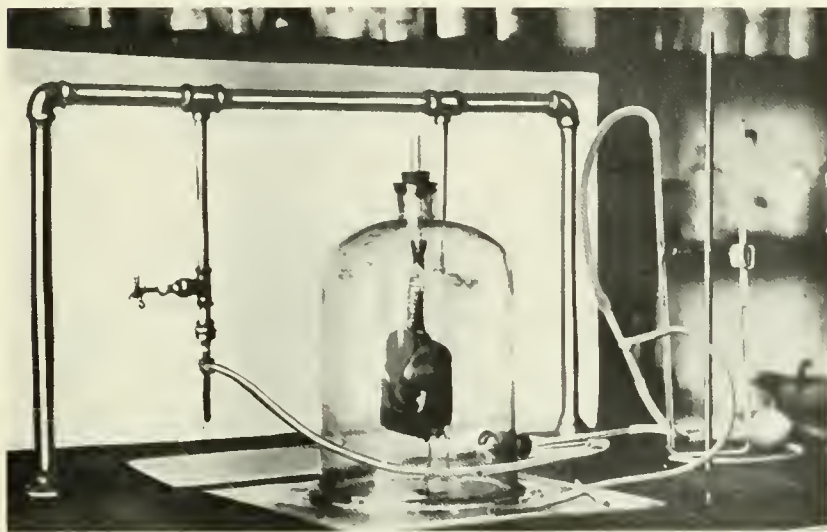


FIG. 7. ARTIFICIAL THORAX USED FOR STUDYING THE FORMATION OF PLEURAL EXUDATES

ditions therefore are ideal for the production of a very low or a markedly negative pressure in the pleural cavity. It would seem reasonable to expect that under such conditions fluid might be sucked out of a "wet" pleura in a manner analogous to the aspiration of fluid out of any very edematous tissue by means of an aspirating syringe. With these ideas in mind the problem was put to an experimental test (37). An artificial thorax (fig. 7) was constructed by means of a bell-jar and a pair of lungs. When the air in the bell-jar around the lungs was aspirated, the lungs would inflate in a manner similar to an

ordinary inspiration. When edematous lungs were used, either those from a case of pneumonia in the human being, or dog's lungs made edematous by pouring into the trachea water containing a little acetic acid, pleural exudate could be seen to pour from the lung surface with each imitated act of respiration. Contrary to the expectation, however, the fluid poured out more rapidly during the act of expiration than during inspiration. The explanation seemed to be that during inspiration the pleura became saturated, and then with the sudden decrease of surface produced by the act of expiration, the fluid was literally squeezed out. So far as I know, this is the only attempt that has been made to study the actual mechanism of the production of pleural exudates. The problem needs more study.

As was stated above on page 420 a resumption of the study in the human of the effects produced in the opposite pleural cavity by injections of air into one showed that when relatively great pressures are reached on one side the response on the other side agrees less closely than is the case when one is dealing with small pressures. Dr. Duff S. Allen and I have recently reinvestigated this question on five human cadavers with normal thoraces (figs. 8 to 12). The experiments were carried out immediately after death, while the bodies were still warm and before rigor mortis had occurred. Two cannulae of the same size were inserted through the sixth interspace into the pleural cavities, one into each side. These were connected with water manometers and also, through a side connection, with an atomizer bulb. By blowing air into one pleural cavity the pressures on both sides could be read simultaneously. It was observed that the pressures in the two pleural cavities agreed most closely when a positive pressure was first created on one side followed in two or three minutes by the injection of air into the opposite side. For example, if a small positive pressure were first created by the injection of air into the right side and then if the left side were gradually inflated by increasing amounts of air, the pressures in the two pleural cavities would agree more closely than when the experiment was carried out in any other way. In fact when dealing with pressures up to about 15 cm. of water the agreement is rather close, as may be seen by reference to the accompanying diagram (fig. 12). The curve of results obtained



FIG. 8. ADULT MALE ONE HOUR AFTER DEATH FROM PERITONITIS

The trachea is in mid-line



FIG. 9. SAME SUBJECT AFTER AIR HAD BEEN INJECTED INTO LEFT PLEURAL CAVITY UNTIL A PRESSURE EQUAL TO THAT OF 8.5 CM. OF WATER WAS OBTAINED

Even with this slight pressure the mediastinal structures are markedly deflected as shown strikingly by the position of the trachea. The left lung seems to have disappeared but instead it is crowded over into the other side.

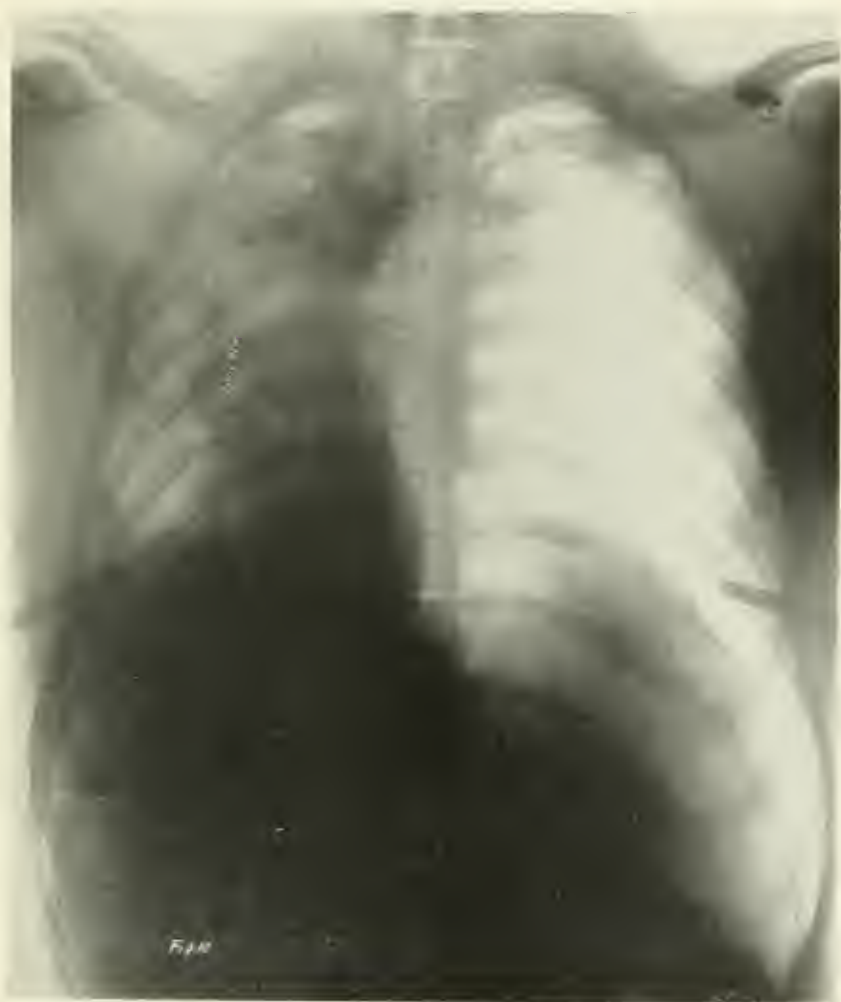


FIG. 10. SAME SUBJECT AFTER INJECTION OF AIR UNTIL A PRESSURE EQUIVALENT TO THAT OF 54 CM. OF WATER WAS OBTAINED

The mediastinal structures are now all on the other side of the mid-line, and there is no evidence of lung tissue on the left side.



FIG. 11. A LIVING NEW-BORN BABY WITH A LEFT PNEUMOTHORAX PRODUCED ACCIDENTALLY IN ATTEMPTS TO INJECT ADRENALIN INTO THE HEART

Note the trachea and heart pushed over into the right side of the chest. The left lung also presumably is pushed over into the right side of the chest instead of being merely "collapsed." Life was compatible with this amount of displacement, although there was a high grade asphyxia until the air in the left chest was aspirated.

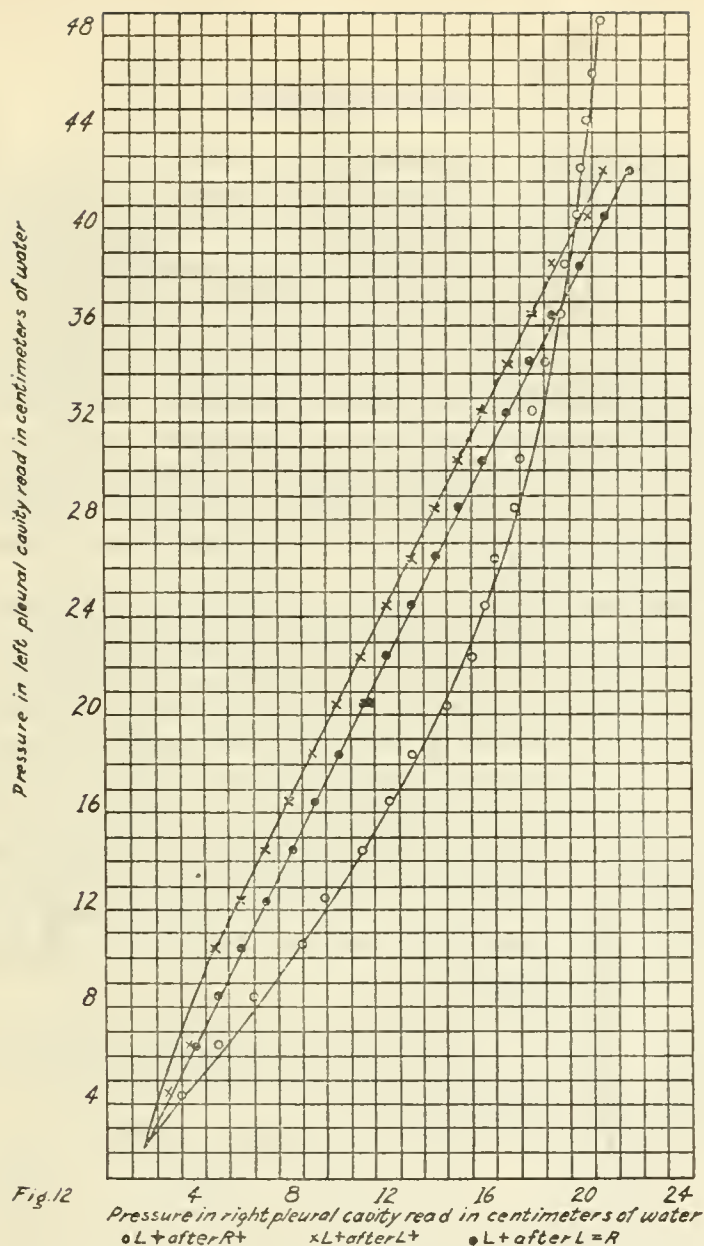


FIG. 12. GRAPHIC REPRESENTATION OF RESULTS OBTAINED WHEN THE PRESSURES IN BOTH PLEURAL CAVITIES ARE REGISTERED AFTER INCREASING THE PRESSURE IN ONE BY THE INJECTION OF AIR

The results shown in this diagram were obtained in an adult male patient one hour after death from generalized peritonitis. If the pressures in the two sides were identical after an alteration in one side, then the plotted graph would form a straight line which would be the oblique diameter of each square through which it passed. The nearest approach to this is the curve marked L+ after R+, a fact which demonstrated that the pressures in the two pleural cavities become most nearly identical if a slight positive pressure with air is made in one side before the other side is inflated.

in the above manner is designated "L+ after R+."³ If, however, the pressures in the two pleural cavities were equal at the start, then continued inflation of one side was not followed by pressure readings in the opposite side of such close agreement, as shown in the curve designated "L+ after L = R." The final one of the three curves, designated "L+ after L+" in figure 12, shows the results obtained when a positive pressure was first created in the left pleural cavity followed by inflation of that cavity in the usual way. The differences obtained in accordance with the actual methods of conducting the experiment probably account for some of the discrepancies and disagreements reported by various observers. Time will not permit a discussion here of the various possible explanations of these differences which suggest themselves.

In this general summary of the effects of alterations of intrapleural pressure which has been given above it is recognized that some of the ideas and theories here set forth may perhaps have to be modified from time to time as later work sheds new light on the subject. At the present time, however, it seems to me that no general theory of the action of pneumothorax explains all the known facts so well as the one which has been advanced above. In principle the theory is the same as that expressed in 1918; it differs only in relatively unimportant details. Those who have disagreed with the principles of the theory have invariably failed to offer a constructive alternative hypothesis which is at all in accord with all the known facts concerning the action of pneumothorax.

³ Under such conditions the numerical values, for example, were found as given in the column below. The close agreement between the pressures of the two sides up to 10 cm. of water is in harmony with our results published in 1918.

2	1.5	14	11.0	26	16.5	38	19.25
4	3.0	16	12.0	28	17.25	40	19.75
6	5.0	18	13.0	30	17.50	42	20.00
8	6.5	20	14.5	32	18.0	44	20.25
10	8.5	22	15.25	34	18.5	46	20.50
12	9.5	24	16.0	36	18.75	48	20.75

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ETIOLOGY AND PREVENTION OF SIMPLE GOITER¹

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INTRODUCTION

Simple or endemic goiter considered from the view point of world medicine is one of the most important and wide spread causes of human suffering and of physical and mental degeneracy with which society has had and still has to deal.

If the thyroid were guilty of all the ills it has been accused of and performed all the functions that have been ascribed to it, it would without doubt be at the same time one of our greatest malefactors as well as one of our greatest benefactors.

I am one of those who believe that the thyroid has very few functions to perform as organ functions go and that we are now in a general way familiar with the most important of these functions. In other words, we probably possess more definite knowledge concerning the anatomy, physiology, chemistry and pathology of the thyroid than of any other gland. On the other hand, as regards the interrelations of the thyroid function with the function of other organs, we are only at the beginning of definite information. Enough is known to recognize that a vast and almost unexplored field of great physiological importance is involved.

Broadly defined, our present conception of the function of the thyroid is that it provides the means, through its iodine containing hormone, of maintaining a higher level of metabolism than would otherwise obtain and also through fluctuations in its activity it provides a means for varying the rate of metabolism to meet changing physiological needs (1, 2).

On the basis of this conception of the normal function of the thyroid the diseases associated with disturbances of its function may be divided into two groups as follows:

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I. Thyroid insufficiencies

1. Simple goiter (endemic, epidemic, sporadic)
2. Myxedema
 - a. Infantile (cretinism)
 - b. Adult (Gull's disease)

II. Exophthalmic goiter.

Reduced to more popular terms these groups would be designated hypo- and hyperthyroidism. They overlap to some extent. Myxedema may supervene in exophthalmic goiter and exophthalmic goiter may develop on the basis of simple goiter although there does not appear to be any necessary relationship between the two groups. This is indicated by the fact that the lower animals are as susceptible to simple goiter as is man; while exophthalmic goiter does not occur spontaneously in them, and up to the present the entire syndrome has not been produced experimentally. Simple goiter is primarily a thyroid disease depending on a deficiency of iodine. Exophthalmic goiter, on the other hand, is primarily dependent upon a constitutional anomaly and the thyroid hyperactivity is of a secondary and compensatory nature. This constitutional anomaly is closely related to that present in *status thymicolymphaticus*, Addison's disease and in experimental *status thymicolymphaticus* produced by sublethal injury of the suprarenal glands. The constitutional anomaly underlying exophthalmic goiter may be congenital or acquired. The acquired form develops most frequently around the period of the menopause, and, in my opinion, explains the increased incidence of exophthalmic goiter at this period of life.

THYROID INSUFFICIENCIES

From the standpoint of clinical medicine, simple goiter and myxedema are usually treated separately but from the standpoint of physiology and pathology they are but different stages or degrees of the same nutritional fault. So far as is known Paracelsus (3) was the first to insist on the close relationship between goiter and cretinism. All subsequent study has strengthened this association which has perhaps been most clearly and briefly expressed by Morel (4) in 1864 as follows: "Goiter is the first stage on the road to cretinism." The same idea has been more comprehensively expressed by Koestl

(5) as follows: "The condition which produces goiter when it is weak also produces cretinism when it is more intense."

The publication of Gull's (6) classical paper in 1874 entitled: "On a cretinoid state supervening in adult life of women" with which he definitely associated atrophy of the thyroid, followed by that of Kocher in 1883 (7) in which he reported 16 cases of complete thyroidectomy in human patients who developed a cachexia similar to that described by Gull, to use Kocher's words, left no doubt of an unmistakable relationship between myxedema and *cachexia strumaprica* on the one hand and cretinism on the other. The final proof of the relation of the thyroid to myxedema was added in 1891 when Murray (8) cured a patient by administering a glycerol extract of sheep's thyroid.

What follows will deal particularly with simple goiter although on account of the association just mentioned infantile myxedema (cretinism) must come into the discussion.

OCCURRENCE AND DISTRIBUTION

Simple goiter includes those thyroid enlargements in man and animals which were formerly grouped under endemic, epidemic, sporadic and physiologic. It may occur in any land and fresh water animal with the ductless thyroid. Animals living in the sea are free from the disease. On the seacoast generally it is ordinarily rare in man and the cases seen are usually in women and in association with pregnancy and lactation. It occurs in all races, in all climates and at all habitable altitudes. In the temperate zones there is a seasonable variation, both in man and animals. It develops more frequently during the late winter and early spring months. Similar seasonal variations also occur in the iodine store of the thyroid as pointed out by Seidell and Fenger (9). While goiter may occur anywhere, even in mid-ocean, as on one of Captain Cook's voyages in 1772, one of the most striking characteristics is the increased incidence in certain more or less defined regions of the world, the so-called districts of endemic goiter (Hirsch) (10). The most notable of these districts are the Himalaya Mountain region of South Central Asia, the Alps, Pyrenees and Carpathian Mountain regions in Europe, the Andean plateau of South America. In North America the most important areas are the

St. Lawrence River and Great Lakes basin, extending west through Minnesota, the Dakotas and the adjacent Canadian provinces and also the Pacific Northwest, including Oregon, Washington and British Columbia. Less important foci occur throughout the Appalachian Mountain region, the Rocky Mountain states and states in the Great Central Basin. It will be noted that most of these regions are mountainous, although there are numerous exceptions. Of greater importance is the occurrence of endemic goiter for the most part on soils deposited from the last glacial period. (See figure 1.)

There is also general evidence of variations in the incidence of goiter in these districts during the last one-hundred years. There are many reports of the sudden occurrence in large numbers of men and animals of goiter, the so-called epidemics. In man these so-called epidemics have for the most part occurred in military garrisons, in institutions and in schools (11, 12). These outbreaks have usually been in districts where the ordinary incidence of goiter is high and in new inhabitants. I have had the opportunity of investigating such outbreaks in dairy herds, on poultry farms and in fish hatcheries (13). Some of these outbreaks were in goiter regions while others were not, showing that with the optimum conditions for its development present, goiter may occur anywhere.

Beginning about the age of puberty, females are more often affected than males. In the districts of severest endemic goiter this difference in incidence due to sex is masked (14). In such regions all of the inhabitants may be affected. In non-goiter regions where only sporadic cases occur these are usually seen in women. Striking an average between these two extremes one may say that in general simple goiter is two or three times more common in females. In the lower animals a difference in incidence due to sex has not been demonstrated. The periods in life when simple goiter most frequently develops are (*a*) during fetal life, (*b*) during pregnancy and lactation, (*c*) during puberty and (*d*) during the menopause.

ETIOLOGY

The cause of thyroid hypertrophy and hyperplasia has interested medical men from the earliest days of medical history. There are few if any diseases in which such a variety of agents have been brought

forward as causal factors. St. Lager in 1867 (15) enumerated over forty theories concerning its causation. Many of these theories must now be grouped as folk-lore and legendary in nature. Poverty, unhygienic living conditions and surroundings, unbalanced diets, certainly are important indirect contributing causes. McCarrison (16) states that it is a common saying in certain provinces of Northern India that people who can afford the luxury of table salt are less likely to be affected. A great variety of organic and inorganic substances both of known and unknown chemical nature occurring in soil and water have been suspected to be the cause. Among these may be mentioned arsenic, salts of calcium and magnesium, sulphides, particularly of iron, fluorides, silica and miasms from decomposing vegetable matter. These substances are not now believed to play any direct rôle. With the development of bacteriology and protozoology the view that thyroid enlargement might be due to a specific living virus was widely adopted. This view has been the basis of extensive investigation. Many types of parasites, bacteria, fungi and protozoa, have been described. McCarrison (17) has worked extensively in this field in India and formerly believed an organism of the colon group was responsible. Infection, he believed, took place through drinking water and food. Chagas (18) working in Brazil in 1909, reported a form of goiter which he thought was due to a trypanosome (*Schizotrypanum cruzi*). Subsequent work has shown that the infection with this organism may exist without thyroid enlargement and that thyroid enlargement may exist without the infection. Kolle (19) of the Swiss Goiter Commission failed to produce thyroid hyperplasia in rats by inoculating them with cultures of this organism. The occurrence of this infection in a district of endemic goiter readily explains the association.

Numerous experiments in which fresh and boiled water from supposedly goiter producing springs were employed have been carried out in recent years by Dieterle, Hirschfeld and Klinger (20), by Bircher (21), by Wilms (22) and by the Swiss Goiter Commission (23). These experiments have yielded either negative or doubtful results. In general nothing suggesting a direct infecting agent has been proven despite the enormous amount of work which has been done. That toxins of bacterial and other origins may indirectly stimulate the thy-

roid to increased activity and possible enlargement is well known. Thyroid enlargements are frequently seen in early pulmonary tuberculosis, in active syphilis, in acute fevers like pneumonia, scarlet fever and diphtheria. The thyroid reaction in these conditions is now explained as dependent upon its increased function in febrile diseases which in turn brings about the increased metabolism occurring in infections and indicates that the thyroid is an element in the defensive mechanism against infection and intoxication.

Water has been associated as the carrier of the active goiter producing agent by all peoples from the remotest times (24). Livingstone has reported that the inhabitants of Central Africa held this belief. Barton (25) stated that the American Indians living in the region of the Great Lakes also believed that water conveyed the causal agent and it is highly improbable that these peoples were familiar with similar views held by the Chinese, the Greeks and the Romans. The conflicting experimental data thus far reported could perhaps be better interpreted as pointing to the lack of some substance (iodin?) necessary for the prevention of thyroid enlargement than that it contained some virus or toxin capable of producing thyroid hyperplasia. My own interest in water as a carrier of the "goiter-ogenic" agent ceased in 1911 when (a) Lenhart and Marine (26) demonstrated that brook trout living in wooden runways (about 30 feet long) between ponds recovered from thyroid hyperplasia although all the trout living in the ponds above and below the runways were highly goiterous and (b) when it was demonstrated that fish with active thyroid hyperplasia put in the tail race below all ponds regularly recovered although living in the most polluted water. If water is a factor in thyroid hyperplasia (and I believe it is) it is due to the absence rather than the presence of some substance. Many years ago we (27) demonstrated that certain species of fish (*Esox*) living in Lake Erie had enlarged thyroids, just as did the land animals of the adjacent country.

Lastly, we will take up the present view of the etiology of simple goiter. This view assumes that goiter is a work or compensatory hypertrophy of the thyroid depending upon a relative or absolute deficiency of iodine. On this basis goiter is only a symptom of a specific deficiency disease in which an insufficient supply of iodine for

one or another cause is the chief factor. The idea that goiter is due to a lack of iodine is not new. Prevost (28) (1830), Maffoni (28), Inglis (28) (1838), Marchand (28), Niepce (28), and particularly Chatin (29) who published the results of his researches in 1852 claimed to have demonstrated a lack of iodine in the air, soil and water in districts of endemic goiter. His claims were attacked and unfortunately so discredited on the grounds of faulty and inadequate methods that further study of this important subject was blocked for forty-five years.

It was not until 1895 when Baumann (30) of Freiburg discovered that iodine in a firm organic combination was a normal constituent of the thyroid, that the possible importance of iodine in thyroid function was revived. He did not succeed in showing that iodine was a necessary constituent although he and his pupils, Roos and Goldmann, (31, 32) were able to show that feeding iodine or food rich in iodine caused a storage of this element in the gland and that in general goiterous thyroids contained less iodine than the normal. Oswald (33, 34, 35) showed that the iodine compound was contained in the globulin or colloid of the gland and that glands rich in colloid usually, though not always, contain more iodine than hyperplastic glands. Further investigations of the nature of this iodine-containing compound resulted in its isolation in 1914 by Kendall (36) in crystalline form and named by him thyroxine.

We may now review in more detail some of the relations which iodine bears to thyroid morphology and function. The relation of iodine to the structure of the thyroid in normal glands, in developing and involuting hyperplasia was first extensively investigated by Marine and Williams (37) and by Marine and Lenhart (38, 39, 40). These studies established the foundation on which the later experimental proof of the fundamental importance of iodine in thyroid physiology and pathology was developed. In comparing the microscopic appearance with the iodine contents of several hundred human and canine goiterous and non-goiterous thyroids, a striking relationship between the iodine store and the histological structure was made out. (See figures 2 and 3.) In the thyroids with normal structure the iodine contents varied between 5.5 and 1 mgm. per gram of dried gland. When the iodine store was found below 0.1 per cent hypertrophic and hyperplastic

changes were regularly found. Comparing further the varying degrees of thyroid hyperplasia with the iodine store of such glands it was found

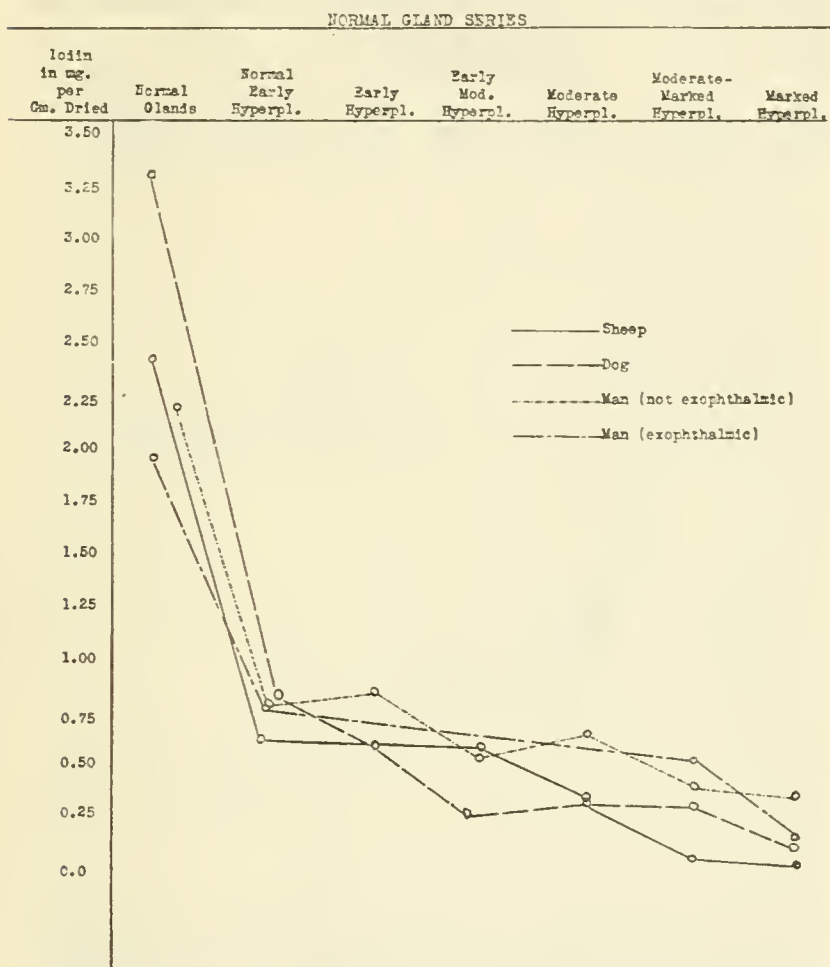


FIG. 2. ILLUSTRATES THE RELATION OF THE IODINE STORE IN SHEEP, DOG, AND HUMAN THYROIDS TO THEIR HISTOLOGICAL STRUCTURE (NORMAL THYROID SERIES)

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that the iodine store progressively decreased as the degree of hyperplasia increased so that in the most marked hyperplasias iodine was absent or present only in traces. A similar relationship between the

iodin store and histological structure of the gland has been observed in large series of ox, pig, sheep, rabbit and fowl thyroids. The generalization may, therefore, be made that the iodine store varies inversely

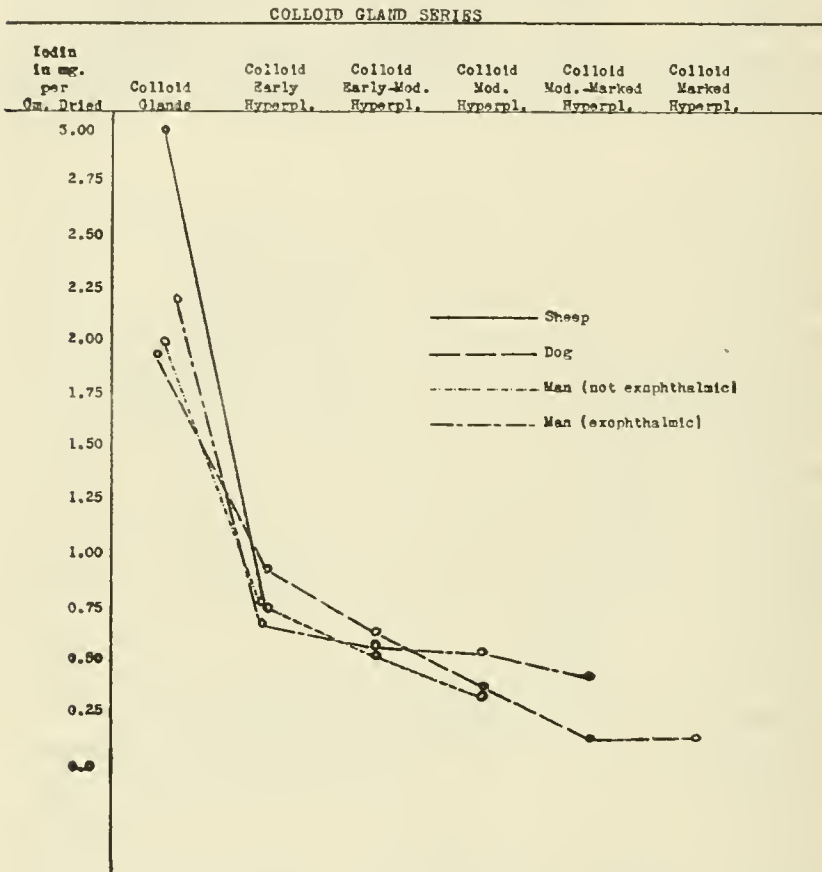


FIG. 3. ILLUSTRATES THE RELATION OF THE IODINE STORE IN SHEEP, DOG, AND HUMAN THYROIDS TO THEIR HISTOLOGICAL STRUCTURE (COLLOID GLAND SERIES)

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with the degree of active hyperplasia and that if the iodine store is higher than 0.1 per cent in these animals no hyperplastic changes are present. This generalization could be readily tested experimentally in several ways:

In the *first* series of experiments iodine was fed to dogs with hyperplastic thyroids after a small piece of the gland had been removed for iodine determination and anatomical classification. Without exception there was found a remarkable storage of iodine in the glands associated with an involution of the hyperplasia to the colloid or resting state (41, 42, 43).

Similar observations have been carried out on rabbits, cats, sheep, fowls and fishes. Complete involution of a marked hyperplasia requires about 15 days in dogs and about forty days in brook trout (44). Sweeping statements in medicine are dangerous but in this instance after many thousand observations I believe it can be stated that iodine will invariably bring about involution of physiological overgrowth of the thyroid. The growth of thyroid carcinoma in both man and dogs is not affected by the administration of iodine nor is iodine retained in active thyroid carcinoma tissue even after many months of feeding (45). The normal thyroid cell has an extraordinary affinity for iodine. This may be demonstrated in both *in vitro* and *in vivo* experiments (46, 47). As much as 18.5 per cent of a single dose of 38 mgm. of iodine given as potassium iodide to a dog by mouth has been recovered from its thyroid whose ratio to body weight was as 1 to 687. Perfused surviving thyroids show that same marked ability to take out and store iodine from the circulating fluid as is seen in the intact gland. Similar perfusions of the kidney and spleen do not result in the retention of iodine (Marine and Feiss).

In a *second* series of experiments Marine and Lenhart (43) studied the effects of the administration and withholding of iodine after partial removal of the thyroid in dogs. As has been shown by many observers, notably by Halsted (48), if one removes from one-half to three-quarters of the gland the remainder undergoes compensatory hyperplasia histologically identical with that seen in the spontaneous hyperplasia of developing goiter.

Large numbers of experiments of partial removal in dogs have been carried out where the histological condition and iodine store were compared and it was found that compensatory hyperplasia did not begin until the iodine store in the remaining part had fallen below 0.1 per cent and that the degree of compensatory hyperplasia increased proportionately as the iodine store decreased just as was found in the spontane-

ous hyperplasias. On the other hand when we removed as much as three-fourths of the gland and administered traces of iodine no compensatory hyperplasia took place in the remaining portion which, as pointed out above, would regularly occur if iodine were withheld. Iodine in any amount will not protect against compensatory hyperplasia when much more than three-fourths of the gland is removed, although the administration of the preformed iodine containing hormone as desiccated thyroid will still further protect the remaining portion against hyperplasia.

A similar effect of iodine may be demonstrated in thyroid transplants in rabbits. As has long been known the thyroid may be readily auto-grafted in any part of the body. Manley and Marine (48) have shown that iodine will prevent the growth and hyperplasia occurring in such transplants or bring about an involution of the hyperplasia if it has occurred irrespective of the location of the transplanted tissue, just as it prevents or causes involution of compensatory hyperplasia occurring in non-transplanted tissues. Such transplants also are capable of storing iodine.

These simple experiments show clearly that the changes in the iodine store and histological structure occurring in experimentally controlled hyperplasia are identical with those observed in spontaneous hyperplasia or simple goiter.

The same general reaction of the thyroid can also be produced in experimental congenital goiter. Halsted (49) first experimentally produced congenital goiter in dogs in 1889. He observed that puppies born from mothers which had had most of the thyroid gland removed had enlarged actively hyperplastic glands at birth—the enlargement reaching twenty times the normal size. He also noted that the histological appearance of such congenital thyroid hyperplasia was identical with the compensatory hyperplasia following partial removal of the gland. The fact that removal of most of the thyroid from the mother ordinarily causes marked enlargement of the fetal thyroid has been confirmed from many sources and for many animals (Edmunds (50), Marine and Lenhart (43), Carlson (51)). In addition to confirming Halsted's observations on dogs, Marine and Lenhart (43) in 1909 added the additional fact that if a few milligrams of iodine are given during pregnancy to dogs from which most of the thyroid has been

removed the young at birth will have normal thyroids, both as regards weight, histological structure and iodine content. We have also been able to obtain from the same animal alternate litters of goiterous and non-goiterous puppies by withholding and administering iodine. This experimental congenital thyroid hyperplasia is true congenital goiter. It is identical with the spontaneous congenital goiter of man and animals and is dependent upon a maternal functional insufficiency of the thyroid. As in the case of many facts of medicine the experimental production of congenital goiter was merely laboratory confirmation of the views held by the older students concerning the etiology of congenital goiter in man. The experimental production and control of congenital goiter should end the discussion in support of the true hereditary nature of congenital goiter.

Another series of experiments was carried out to demonstrate that the normal thyroid could be kept normal over long periods of time even in highly goiterous districts by the administration of small amounts of iodine. In these experiments litters of puppies from iodized mothers were used to insure normal thyroids at birth. To half of each litter one milligram of iodine was given by mouth each week while the other half of each litter served as controls. These experiments were carried out in Cleveland where practically all dogs normally have some degree of thyroid hyperplasia. It was found that those puppies which did not receive iodine developed the usual thyroid enlargement while the thyroids of those that received a milligram once a week remained normal although both control and iodine fed puppies were kept in the same kennels and fed with the same kind of food. These experiments proved that iodine in minute amount could control thyroid overgrowth and that we had to deal with a unique physiological fact in that a single inorganic element determined the functional value of the thyroid secretion.

Diet plays a part in the etiology of thyroid overgrowth (goiter) (52). St. Lager (15) (1867) stated that foods rich in fats—particularly pork had long been considered as an important factor in the cause of goiter. Baumann and his pupils noted that foods rich in iodine (codfish) caused a storage of iodine in the gland and that fresh meats caused a decrease in the iodine store. Watson (53) also found that meat diets caused hypertrophy and hyperplasia of the rat's thyroid.

Marine and Lenhart showed that pig's liver was the most potent of a great variety of meats in causing thyroid hyperplasia in dogs. We also found that this food was an important factor in the causation of thyroid overgrowth in brook trout, a disease which at that time (1910) was widespread throughout the fish hatcheries in the United States where the Salmonidae were artificially reared (13, 26).

In investigating an acute outbreak of goiter in poultry in the spring of 1914 the high protein and fat content (meat scrap) of the diet was believed to be an important factor in causing thyroid overgrowth. We also noted in a herd of dairy cattle in which several goiterous and cretin calves were born that the high proportion of cotton seed meal used in the food was probably an important factor. McCarrison (54) has shown that diets which include an excessive amount of digestible fats free from iodine (lard and butter) were even more potent in producing thyroid hyperplasia in pigeons than were the high protein and fat diets above mentioned. These observations on the effects of fat feeding have been confirmed by Mellanby (55). McCarrison further showed that fats rich in iodine, as codliver oil, did not cause thyroid hyperplasia. This probably explains why goiter is rare among the Eskimos in spite of their high fat and protein diets. Shapiro and Marine (56) (unpublished) confirmed McCarrison's observation by showing that the administration of 10 cc. of cottonseed oil daily to rabbits in addition to their ordinary diet regularly causes a striking thyroid overgrowth, just as the diet of pig's liver did in brook trout. It usually takes from sixty to ninety days to produce marked thyroid enlargement in rabbits. As thyroid hyperplasia is secondary to the depletion of the iodine store these facts indicate that diets rich in proteins and fats increase the rate of discharge of iodine. It seems probable that thyroid activity is more necessary for the metabolism of fats and proteins than of carbohydrates. Carbohydrate diets as shown by McCarrison do not cause thyroid hyperplasia.

Lastly, I will call attention to the influence of *pregnancy and lactation* in the etiology of thyroid enlargement. The period of pregnancy and lactation is one of the three important periods in life when simple goiter most frequently develops. The association has been known since antiquity. Lawson Tait (57) in 1875 described the step like enlargement of the thyroid associated with multiple pregnancies.

Slight hypertrophy of the thyroid during pregnancy often to the stage of clinical detectability is looked upon by obstetricians as normal even in non-goiterous districts. The physiological nature of the enlargement has not been extensively investigated. Naturally one would suspect from what we know of normal thyroid physiology that the hypertrophy during pregnancy was dependent upon increased demands for functional activity during this period. Murlin (58) in his studies on metabolism during pregnancy, both in human cases and in dogs, observed only a slight increase in the total energy production, averaging about 4 per cent in the case of woman and 9 per cent in a dog which was proportional to the increase in the weight of the fetus. Baer, (59) in 45 normal human cases, found the basal metabolic rate in late pregnancy to average about 30 per cent above the normal for non-pregnant women, of approximately similar surface areas. Root and Root (60) in a careful study of human pregnancy found an increase in the basal metabolic rate beginning at the fifth month and reaching + 23 per cent eleven days before delivery. During the last two years, we have studied the influence of pregnancy in rabbits, with and without thyroidectomy, on heat production (61). Our results show that heat production markedly increases in this animal during pregnancy and lactation. The rise begins about the middle of pregnancy, that is about the end of the second week of gestation, and continues through the period of lactation. Thyroidectomy performed during the first week of pregnancy greatly reduces and sometimes prevents the increased heat production both of pregnancy and lactation. Thyroidectomized rabbits occasionally abort but we have never seen an instance of abnormally prolonged pregnancy after thyroidectomy as has been described. A large percentage of these rabbits abandon their young at birth. These experiments, we believe, indicate that normally there is a greater increase in heat production during pregnancy than some of the previous work has indicated, and that this increase is in part dependent upon the maternal thyroid and that the enlargement which may occur at this time is essentially a compensatory or work hypertrophy. The enlargement of the fetal thyroid, which is so directly dependent upon maternal thyroid insufficiency is also, we believe, a purely physiological or compensatory reaction.

To sum up the evidence at present available concerning the etiology of goiter, I believe we may abandon the older views that it is due to a specific living virus or a specific chemical substance and conclude that simple goiter is a compensatory or work hypertrophy depending upon a variety of metabolic stimuli for functional activity of the thyroid which depletes its iodine store. The immediate cause of thyroid enlargement is a relative or an absolute deficiency of iodine. This deficiency may result from:

1. Any factors which increase the needs of the organism for the iodine containing hormone as occur during puberty, pregnancy and lactation, during the menopause, during certain infections and intoxications, following sufficient injury to the interrenal gland, or as a result of diets consisting mainly of fat and protein.

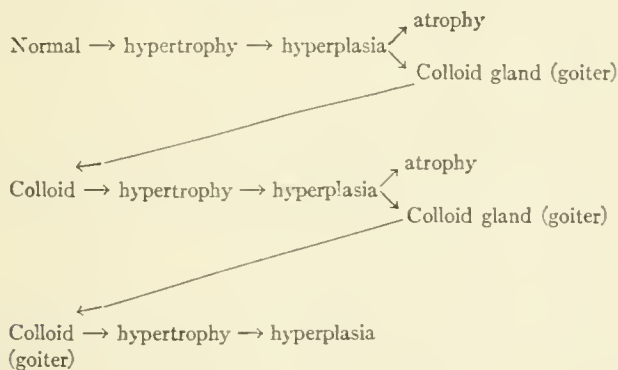
2. Any factors which interfere with the absorption or utilization of the normal intake of iodine. We have no knowledge as yet of such factors although it is conceivable that the intestinal bacterial flora or intestinal parasites could utilize or divert part of the iodine intake.

3. Factors which bring about an abnormally low intake or actual deprivation of iodine, either natural or experimental. The normal source of iodine is from food and water though traces may be taken by breathing the air in the immediate vicinity of the sea. In districts of endemic goiter both the food and water derived from such soils have been proven to be very low in iodine (29), Olin (62), McClendon (63).

When one recalls that the maximum store of iodine in the normal thyroid is about 25 mgm. and that an intake of 50 mgm. if properly distributed may maintain the thyroid in a normal state for as long as a year, that certain diets, pregnancy, fevers and other conditions bringing about increased metabolism can quickly exhaust the iodine store of the thyroid, it would appear that the iodine deficiency might be considered as a primary and possibly the essential cause of goiter. Nevertheless, we must still consider the possibility that some chemical agent or toxin may be operating in districts of endemic goiter to divert an otherwise adequate intake of iodine or to increase the needs of the organism for thyroid activity, and not rest content merely with the fact that for practical purposes thyroid hyperplasia is due to iodine deficiency.

ANATOMICAL CYCLE AND PATHOLOGICAL ANATOMY

The thyroid is a very labile tissue capable of marked and rapid hyperplasia and involution in response to variations in functional activity. On this account a wide range and a great variety of progressive, regressive, degenerative, inflammatory, atrophic and neoplastic changes may be present, many of which as Virchow (64) pointed out are only terminal metamorphoses and complications in long standing goiters. These secondary changes are usually present in human goiter and have caused most of the confusion and difficulties of interpretation. The primary and essential anatomical changes in the thyroid in goiter we believe are relatively simple (65). As a result of the study of large series of goiterous and non-goiterous thyroids in fish, birds, dogs, sheep, oxen and man it has been found that the cycle of essential cell changes may be reduced to the following scheme:



We believe this is the only cycle of cell change of which the thyroid tissue is capable and that it tends to undergo this cycle in response to stimuli requiring increased thyroid activity.

The gland cells begin to hypertrophy when the iodine store falls below a given level (less than 0.1 per cent in the animals studied) and continue this hypertrophy and hyperplasia until exhaustion atrophy or recovery supervenes. By anatomical recovery one means the involution of the active hyperplasia to the colloid or resting stage and not as some have supposed, the disappearance of the thyroid enlargement. Colloid goiter is the nearest condition to the normal both anatomically and chemically that a thyroid gland which has once

been in the state of active hyperplasia can assume and such colloid glands are capable of manifesting all of the reactions which a normal gland can show (42). Thus they can repeat the cycle of hypertrophy, hyperplasia and involution many times during the life history of the animal. The step-like enlargements seen in association with multiple pregnancies is a clinical manifestation of alternating hyperplasia and involution. This has been shown experimentally in dogs by producing compensatory hyperplasia by partial removal of the gland, then causing involution of this hyperplasia with iodine; again producing compensatory hyperplasia by partial removal and again producing involution with iodine. We have succeeded by this means in causing the gland to repeat its cell cycle as many as seven times in the same animal during a period of eighteen months.

These cell changes are not specific for any clinical disease (66, 67) as some have supposed but occur in response to any stimulus for increased thyroid activity which brings about a sufficient depletion of the iodine store.

The first change in the thyroid in developing goiter is a marked decrease in the iodine store. When this has fallen below 0.1 per cent increased vascularity, cell hypertrophy and hyperplasia occur. The stainable colloid decreases as the hyperplasia increases. In the lower animals, with the exception of the rat, this hyperplasia of the epithelium is regular and uniform while in man it is frequently irregular and nodular—so-called struma nodosa or adenomatous goiter (68). These nodules or adenomas are due to different rates of growth of foci of thyroid cells in response to a general stimulus to the thyroid. It is believed they may arise both from differentiated thyroid alveoli and from cell rests as first described by Wölfler (69). These adenomas are an integral and essential part of endemic goiter in man and are due to the same stimulus which excites the thyroid as a whole to hypertrophy. These multiple circumscribed benign growths are functionally active yet have certain of the attributes of tumor, one of which is that their growth once initiated is frequently not controlled by iodine as is every simple hyperplasia (70). The terminal metamorphoses are far more serious than those of simple goiter. Since in addition to pressure effects, hemorrhage, necrosis, cyst formation and their possible rôle in the etiology of Graves' disease, probably 90 per cent of the malignant tumors of the thyroid arise from them.

PREVENTION

The curative treatment of simple goiter and cretinism has been actively practised for some twenty-five hundred years without any notable decrease in the incidence. Nor has the introduction of surgery during the last fifty years materially affected the general problem of control.

There is some general evidence that the severer degrees of goiter and cretinism are less common than formerly and also that certain regions where mild endemic goiter formerly existed are now relatively free. It is possible that this result is an indirect effect of the general economic betterment—higher standards of living, improved sanitation, a better and larger variety of food, control of infectious and contagious diseases and more extensive means of transportation. Some more direct method of prevention must be developed if we are to control the disease. The search for such methods is not new. This was one of the main purposes of the Goiter Commissions appointed by the Sardinian government in 1845, by the Austrian government in 1860, by the French government in 1863, by the Swiss government in 1908, and more recently by the Italian government. The installation of new water supplies was recommended by some of the goiter commissions. In certain instances this is said to have decreased and in others to have increased the incidence. In Vienna, Austria, Portland, Oregon, and Seattle the new water supplies even though exceptionally low in bacteria, organic and inorganic matter seem to have resulted in an increase in goiter. Changing the water supply for the purpose of goiter prevention is at present without scientific support. As a result of the experimental data regarding the relation of iodine to thyroid physiology and pathology and the conclusion drawn from them, that simple goiter was due to a relative or absolute deficiency of iodine, the plan of supplying a minute amount of iodine to meet this deficiency suggested itself.

The first occasion when iodine was thus used in the prevention of simple goiter on a large scale was in 1909 and 1910 (13, 26) when Marine and Lenhart demonstrated that this substance added to the water supply in a concentration not greater than 1 mgm. per liter arrested and prevented the development of thyroid hyperplasia in

brook trout. It was later shown that changing the diet of the fish to whole sea-fish which also contained traces of iodine was equally efficacious (71). Since then the administration of iodine in some form or manner has been successfully applied in the prevention of goiter in sheep, cattle, pigs and poultry (72, 73, 74). Attempts to apply the plan of goiter prevention in man on a large scale was beset with difficulties, obstacles and hostility which society seems always to have interposed when new methods of controlling disease (however meritorious) have been proposed. Many interesting stories could be told of our efforts and failures to make the experiment.

The prevention of simple goiter in man was not attempted on any large practical scale until 1917 (75) when O. P. Kimball and I began its practical application among the girls attending the public schools of the city of Akron, Ohio. The method used at that time consisted in the administration of 2 grams of sodium iodide in 0.2 gram doses distributed over a period of two weeks and repeated each autumn and spring. At that time it was pointed out that this amount of iodine was excessive and far beyond the needs of the individual or the ability of the thyroid to utilize or store it. This demonstration was carried out for a period of two and one-half years and the results are briefly as follows: Of 2190 pupils taking 2 grams of sodium iodide twice yearly only five developed thyroid enlargement while of 2305 girls not taking the prophylactic 495 developed thyroid enlargement. The thyroid glands of 773 pupils out of 1182 with thyroid enlargement at the first examination who took the prophylactic treatment have decreased in size; while in 145 out of 1049 pupils with thyroid enlargement at the first examination who did not take the prophylactic treatment the thyroids have decreased in size (76).

These figures demonstrate in a striking manner both the preventive and the curative effects of minute doses of iodine. They are just as striking as the results obtained in animal experiments and the generalization may be made that human goiter is as easily and as cheaply prevented as it is in the lower animals. Since these results were reported numerous papers have appeared in the European literature particularly from Switzerland and Italy, confirming these observations and extending the prophylactic use of iodine. Klinger (77) in 1921 reported even more striking curative results in the school children of

Zürich. He worked with a school population in which the incidence of goiter varied from 82 to 95 per cent while our maximum incidence in Akron was 56 per cent.

Thus, of 760 children 90 per cent were goiterous at the first examination. After fifteen months treatment with 10 to 15 mgm. iodine weekly only 28.3 per cent were goiterous of a total of 643 reexamined. Independent reports by Fritsche (78), Bayard (79), Hunziker and Wyss (80), and deQuervain (81) from different Swiss towns indicate equally beneficial results. Similar reports from Italy have been made by Muggia (82) and Pighini (83) during the last two years. In 1922 the Swiss Goiter Commission (84) recommended the introduction of state wide prophylactic measures against endemic goiter in either of two ways:

1. For general prophylaxis the use of salt containing 2 to 5 mgm. potassium iodide per kilogram.
2. For school prophylaxis the use of tablets containing 1 mgm. iodine at weekly intervals.

These amounts of iodine are too small.

Iodine is effective when administered in any form or by any means: by inhalation, by mouth, by application to the skin or by injection. This fact introduces difficulties and advantages—difficulties as regards the selection of the best form and means of administration, and advantages in that the desired results may be accomplished with certainty in a variety of ways. The ideal plan of administration is still to be worked out. Historically (85, 86, 87) and of course quite unknowingly iodine containing salt was the first means of administration and I am of the opinion that ultimately it will prove the best means of administration when the entire population is to be protected. Sea salt or a salt containing 2 to 5 mgm. of iodine per kilo, if used exclusively would seem ample in the districts where endemic goiter is not very prevalent. If applied to the school population only, the Swiss plan of giving tablets containing 5 to 10 mgm. at weekly intervals is more convenient and quite sufficient for prevention. For curative effects larger amounts will probably be necessary. Simple goiter most frequently develops (*a*) during fetal life, (*b*) during adolescence, and (*c*) during pregnancy and lactation. Any plan of prevention that controls thyroid growth during these

three periods would practically eliminate simple goiter. Goiter in the mother and fetus can be prevented as easily and as simply as that of adolescence and by the same means. All pregnant women living in goiter districts should be given the equivalent of 10 mgm. of iodine each week during the period of pregnancy and lactation.

The dangers and untoward effects of iodine when used in the minimum amounts suggested have been exaggerated. Iodine like other foods has been greatly abused and its abuse in the prevention and treatment of goiter will continue until it is realized that the maximum storage capacity of the normal thyroid for iodine is not over 25 mgm. and that such a store is sufficient to meet the ordinary physiological needs of the organism for months. When one considers the extent to which iodine is daily used both by the profession and the public the probability of injury becomes negligible. Iodism and the possibility of aggravating incipient exophthalmic goiter are the only dangers worthy of consideration. In general iodine should not be used in Graves' disease although there are unquestionably stages of the disease in which daily doses of 1 mgm. may be helpful.

Desiccated thyroid is theoretically a better prophylactic than iodine but practically it is too dangerous a drug to be recommended at present.

Cretinism: Prevention offers the only certain means of controlling endemic cretinism both in man and in animals. In sharp contrast with Gull's disease postnatal treatment is only partially successful because too much damage has been done before treatment can be instituted. Remarkable cures may be obtained in cretin animals when treatment can be started shortly after birth (88). Since all available evidence indicates that endemic cretinism is due to the same nutritional fault as endemic goiter and since congenital myxedema in animals may be so easily controlled by the administration of iodine to the mother during pregnancy, I believe the prevention of endemic cretinism in man is as simple as the prevention of simple goiter and can be accomplished by the same means.

Simple goiter is the easiest and cheapest of all known diseases to prevent and its control may be accomplished by available methods as soon as organized society determines to make the effort. The prevention of goiter will mean a great deal more than eliminating this form

of cervical deformity. It means in addition the control of those forms of physical and mental degeneracy such as cretinism, mutism and idiocy, which are dependent upon thyroid insufficiency. Further, it would prevent the development of thyroid adenomas. The terminal metamorphosis of adenomas are far more serious than those of simple hyperplasia since in addition to hemorrhage and cyst formation probably 90 per cent of the malignant tumors of the thyroid arise from these growths. Many investigators have likened the effects of thyroidectomy and the manifestations of cretinism to premature senility. Arteriosclerosis commonly occurs in thyroidectomized lambs (Simpson) (89), in myxedema (of adults) and in endemic myxedema (cretinism). It has also been pointed out that carcinomas in general appear earlier in life in highly goiterous districts than in nongoiterous districts. The elimination of endemic goiter would eliminate this particular form of premature senility and the sequelae which it entails.

In conclusion one can say that the factors which cause simple goiter center about the supply of iodine and the needs, normal and abnormal, of the thyroid gland for iodine. Supplying this element in an amount that roughly could be considered as approximating the physiological demand has resulted in preventing simple goiter in both man and animals and in preventing cretinism in animals.

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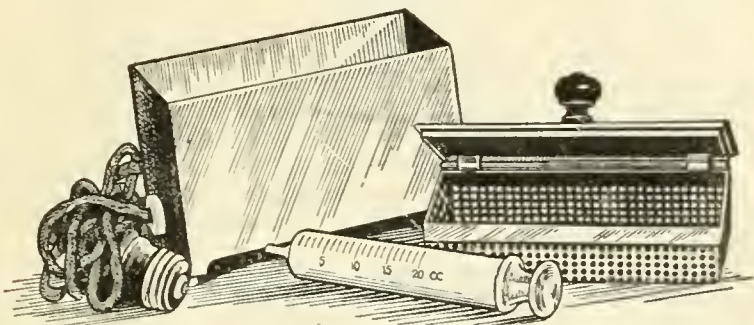
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